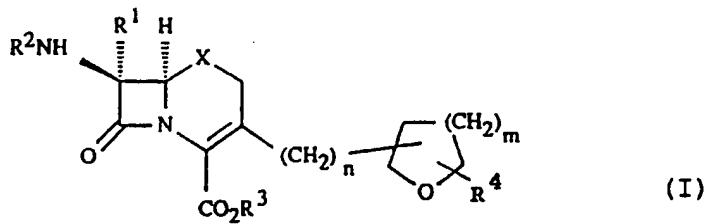




## INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(51) International Patent Classification 5 : C07D 501/20, 501/18, 463/00 A61K 31/545, 31/435		A1	(11) International Publication Number: <b>WO 92/01696</b> (43) International Publication Date: 6 February 1992 (06.02.92)
(21) International Application Number: PCT/GB91/01228 (22) International Filing Date: 22 July 1991 (22.07.91)		(74) Agent: WHITE, Susan, Mary; SmithKline Beecham, Corporate Patents, Great Burgh, Yew Tree Bottom Road, Epsom, Surrey KT18 5XQ (GB).	
(30) Priority data: 9016189.4 24 July 1990 (24.07.90) GB 9109540.6 2 May 1991 (02.05.91) GB		(81) Designated States: AT (European patent), AU, BE (European patent), CA, CH (European patent), CS, DE (European patent), DK (European patent), ES (European patent), FI, FR (European patent), GB (European patent), GR (European patent), HU, IT (European patent), JP, KR, LU (European patent), NL (European patent), NO, PL, SE (European patent), US.	
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		Published <i>With international search report. Before the expiration of the time limit for amending the claims and to be republished in the event of the receipt of amendments.</i>	

(54) Title: CEPHALOSPORINS AND HOMOLOGUES, PREPARATIONS AND PHARMACEUTICAL COMPOSITIONS



## (57) Abstract

$\beta$ -Lactam antibiotics of formula (I) or a salt thereof, wherein R<sup>1</sup> is hydrogen, methoxy or formamido; R<sup>2</sup> is an acyl group; CO<sub>2</sub>R<sup>3</sup> is a carboxy group or a carboxylate anion, or R<sup>3</sup> is a readily removable carboxy protecting group; R<sup>4</sup> represents up to four substituents; X is S, SO, SO<sub>2</sub>, O or CH<sub>2</sub>; m is 1 or 2; and n is 0, useful in the treatment of bacterial infections in humans and animals.

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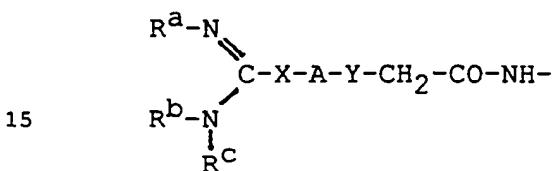
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## Cephalosporins and homologues, preparations and pharmaceutical compositions

This invention relates to novel  $\beta$ -lactam containing compounds, their preparation and their use, and in particular to a novel class of cephalosporins. These compounds have antibacterial properties, and are therefore of use in the treatment of bacterial infections in humans and animals caused by a wide range of organisms.

10 GB 1 385 831 (Hoechst) claims 7-acylamino-cephem-carboxylic acid compounds substituted at the 7-position by a group:



in which  $\text{R}^a$  and  $\text{R}^b$ , which may be the same or different, each represents a hydrogen atom or an alkyl group having from 1 to 5 carbon atoms or  $\text{R}^a$  and  $\text{R}^b$  together represent an alkylene group which may be substituted,  $\text{R}^c$  represents a hydrogen atom or an alkyl group having from 1 to 5 carbon atoms,  $\text{X}$  represents a single bond or an  $\text{NH}$  group,  $\text{A}$  represents a phenylene or thiophenylene group which may be substituted and  $\text{Y}$  represents a single bond or an oxygen atom;

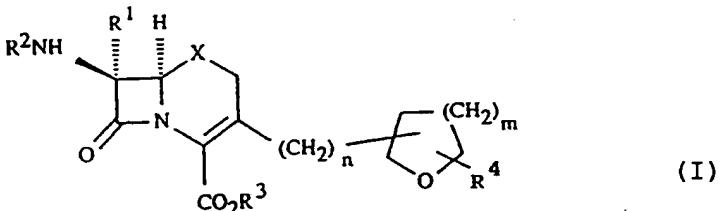
and substituted at the 3-position by an alkyl group having from 1 to 5 carbon atoms, or a cyclo-alkyl group having from 3 to 7 ring carbon atoms which may include one or more hetero ring atoms. Tetrahydrofuran is described as an example of a 3-position substituent from a list of 14 radicals. The Examples describe only methyl, ethyl and isopropyl groups at the 3-position of the cephalosporin nucleus.

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We have now found a particular class of cephalosporins bearing a cyclic ether substituent at the 3-position of the cephalosporin nucleus that possesses prolonged and high levels of antibacterial activity, and shows good absorption both parentally and orally, especially orally.

The present invention provides a compound of formula (I) or a salt thereof:

10



15

wherein

$\text{R}^1$  is hydrogen, methoxy or formamido;

$\text{R}^2$  is an acyl group, in particular that of an 20 antibacterially active cephalosporin;

$\text{CO}_2\text{R}^3$  is a carboxy group or a carboxylate anion, or  $\text{R}^3$  is a readily removable carboxy protecting group (such as a pharmaceutically acceptable in vivo hydrolysable ester group);  $\text{R}^4$  represents up to four substituents selected from 25 alkyl, alkenyl, alkynyl, alkoxy, hydroxy, halogen, amino, alkylamino, acylamino, dialkylamino,  $\text{CO}_2\text{R}$ ,  $\text{CONR}_2$ ,  $\text{SO}_2\text{NR}_2$  (where  $\text{R}$  is hydrogen or  $\text{C}_{1-6}$  alkyl), aryl and heterocyclyl, which may be the same or different and wherein any  $\text{R}^4$  alkyl substituent is optionally substituted by any other  $\text{R}^4$  substituent;  $\text{X}$  is  $\text{S}, \text{SO}, \text{SO}_2, \text{O}$  or  $\text{CH}_2$ ;  $\text{m}$  is 1 or 2; and  $\text{n}$  is 30 0.

The bonding carbon atom of the cyclic ether moiety which links the ring to the cephalosporin nucleus is generally 35 asymmetric. The present invention includes either

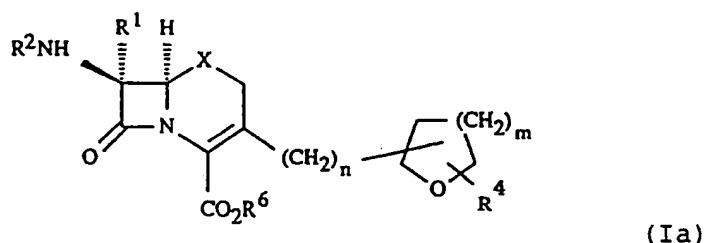
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stereoisomer, as well as mixtures of both isomers.

In compounds of formula (I) wherein R<sup>1</sup> is formamido, the formamido group can exist in conformations wherein the 5 hydrogen atoms of the -NH-CHO moiety are cis- or trans-; of these the cis conformation normally predominates.

Since the  $\beta$ -lactam antibiotic compounds of the present invention are intended for use as therapeutic agents in 10 pharmaceutical compositions, it will be readily appreciated that preferred compounds within formula (I) are pharmaceutically acceptable, i.e. are compounds of formula (Ia) or pharmaceutically acceptable salts or pharmaceutically acceptable in vivo hydrolysable esters 15 thereof:

20



wherein R<sup>1</sup>, R<sup>2</sup>, R<sup>4</sup>, m, n and X are as defined with respect to formula (I) and the group CO<sub>2</sub>R<sup>6</sup> is CO<sub>2</sub>R<sup>3</sup> where CO<sub>2</sub>R<sup>3</sup> is a 25 carboxy group or a carboxylate anion.

Accordingly, the present invention provides a compound of formula (Ia) or a pharmaceutically acceptable salt or in vivo hydrolysable ester thereof, for use as a therapeutic 30 agent, and in particular an in vivo hydrolysable ester thereof for use as an orally administrable therapeutic agent.

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The present invention further provides a compound of formula (Ia) or a pharmaceutically acceptable salt or in vivo hydrolysable ester thereof, for use in the treatment of bacterial infections, more particularly an in vivo hydrolysable ester thereof for use in the oral treatment of bacterial infections.

The present invention also includes a method of treating bacterial infections in humans and animals which comprises 10 the administration of a therapeutically effective amount of an antibiotic compound of this invention of the formula (Ia) or a pharmaceutically acceptable in vivo hydrolysable ester thereof, in particular the oral administration of a therapeutically effective amount of an in vivo hydrolysable 15 ester.

In addition, the present invention includes the use of a compound of formula (Ia) or a pharmaceutically acceptable salt or in vivo hydrolysable ester thereof, for the 20 manufacture of a medicament for the treatment of bacterial infections, in particular the use of an in vivo hydrolysable ester for the manufacture of a medicament for the oral treatment of bacterial infections.

25 Those compounds of the formula (I) wherein R<sup>3</sup> is a readily removable carboxy protecting group other than a pharmaceutically acceptable in vivo hydrolysable ester or which are in non-pharmaceutically acceptable salt form are primarily useful as intermediates in the preparation of 30 compounds of the formula (Ia) or a pharmaceutically acceptable salt or pharmaceutically acceptable in vivo hydrolysable ester thereof.

Suitable readily removable carboxy protecting groups for the 35 group R<sup>3</sup> include groups forming ester derivatives of the carboxylic acid, including in vivo hydrolysable esters. The derivative is preferably one which may readily be cleaved in vivo.

It will be appreciated that also included within the scope of the invention are salts and carboxy-protected derivatives, including in vivo hydrolysable esters, of any 5 carboxy groups that may be present as optional substituents in compounds of formula (I) or (Ia). Also included within the scope of the invention are acid addition salts of any amino group or substituted amino group that may be present as optional substituents in compounds of formula (I) or 10 (Ia).

Suitable ester-forming carboxyl-protecting groups are those which may be removed under conventional conditions. Such groups for R<sup>3</sup> include benzyl, p-methoxybenzyl, 15 benzyloxymethyl, p-nitrobenzyl, 4-pyridylmethyl, 2,2,2-trichloroethyl, 2,2,2-tribromoethyl, t-butyl, t-amyl, allyl, diphenylmethyl, triphenylmethyl, adamanyl, 2-benzyloxyphenyl, 4-methylthiophenyl, tetrahydrofuran-2-yl, tetrahydropyran-2-yl, pentachlorophenyl, acetonyl, 20 p-toluenesulphonyl, methoxymethyl, a silyl, stannyl or phosphorus- containing group, an oxime radical of formula -N=CHR<sup>7</sup> where R<sup>7</sup> is aryl or heterocyclic, or an in vivo hydrolysable ester radical such as defined below.

25 When used herein the term 'aryl' includes phenyl and naphthyl, each optionally substituted with up to five, preferably up to three, groups selected from halogen, mercapto, C<sub>1-6</sub> alkyl, phenyl, C<sub>1-6</sub> alkoxy, hydroxy(C<sub>1-6</sub>)alkyl, mercapto(C<sub>1-6</sub>)alkyl, halo(C<sub>1-6</sub>) alkyl, 30 hydroxy, amino, nitro, carboxy, C<sub>1-6</sub> alkylcarbonyloxy, alkoxy carbonyl, formyl, or C<sub>1-6</sub> alkylcarbonyl groups.

The terms 'heterocyclyl' and 'heterocyclic' as used herein include aromatic and non-aromatic, single and fused, rings 35 suitably containing up to four hetero-atoms in each ring selected from oxygen, nitrogen and sulphur, which rings may be unsubstituted or substituted by, for example, up to three

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groups selected from halogen, (C<sub>1-6</sub>)alkyl, (C<sub>1-6</sub>)alkoxy, halo(C<sub>1-6</sub>)alkyl, hydroxy, carboxy, carboxy salts, carboxy esters such as (C<sub>1-6</sub>)alkoxycarbonyl, (C<sub>1-6</sub>)alkoxycarbonyl(C<sub>1-6</sub>)alkyl, aryl, and oxo groups. Each 5 heterocyclic ring suitably has from 4 to 7, preferably 5 or 6, ring atoms. The term 'heteroaryl' refers to heteroaromatic heterocyclic rings. A fused heterocyclic ring system may include carbocyclic rings and need include only one heterocyclic ring. Compounds within the invention 10 containing a heterocyclyl group may occur in two or more tautomeric forms depending on the nature of the heterocyclyl group; all such tautomeric forms are included within the scope of the invention.

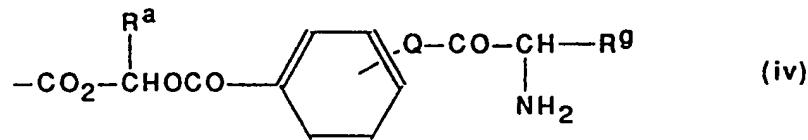
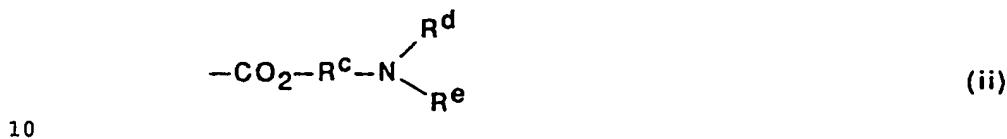
15 When used herein the terms 'alkyl' alkenyl, alkynyl and 'alkoxy' include straight and branched chain groups containing from 1 to 6 carbon atoms, such as methyl, ethyl, propyl and butyl. A particular alkyl group is methyl.

20 When used herein the term 'halogen' refers to fluorine, chlorine, bromine and iodine.

A carboxyl group may be regenerated from any of the above esters by usual methods appropriate to the particular R<sup>3</sup> 25 group, for example, acid- and base- catalysed hydrolysis, or by enzymically-catalysed hydrolysis, or by hydrogenolysis under conditions wherein the remainder of the molecule is substantially unaffected.

30 Examples of suitable pharmaceutically acceptable in vivo hydrolysable ester groups include those which break down readily in the human body to leave the parent acid or its salt. Suitable ester groups of this type include those of part formulae (i), (ii), (iii), (iv) and (v):

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wherein  $\text{R}^{\text{a}}$  is hydrogen,  $\text{C}_{1-6}$  alkyl,  $\text{C}_{3-7}$  cycloalkyl, methyl, or phenyl,  $\text{R}^{\text{b}}$  is  $\text{C}_{1-6}$  alkyl,  $\text{C}_{1-6}$  alkoxy, phenyl, benzyl,  $\text{C}_{3-7}$  cycloalkyl,  $\text{C}_{3-7}$  cycloalkyloxy,  $\text{C}_{1-6}$  alkyl  $\text{C}_{3-7}$  cycloalkyl, 1-amino  $\text{C}_{1-6}$  alkyl, or 1-( $\text{C}_{1-6}$  alkyl)amino  $\text{C}_{1-6}$  alkyl; or  $\text{R}^{\text{a}}$  and  $\text{R}^{\text{b}}$  together form a 1,2-phenylene group optionally substituted by one or two methoxy groups;  $\text{R}^{\text{c}}$  represents  $\text{C}_{1-6}$  alkylene optionally substituted with a methyl or ethyl group and  $\text{R}^{\text{d}}$  and  $\text{R}^{\text{e}}$  independently represent

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$C_{1-6}$  alkyl;  $R^f$  represents  $C_{1-6}$  alkyl;  $R^g$  represents hydrogen or phenyl optionally substituted by up to three groups selected from halogen,  $C_{1-6}$  alkyl, or  $C_{1-6}$  alkoxy; Q is oxygen or NH;  $R^h$  is hydrogen or  $C_{1-6}$  alkyl;  $R^i$  is hydrogen, 5  $C_{1-6}$  alkyl optionally substituted by halogen,  $C_{2-6}$  alkenyl,  $C_{1-6}$  alkoxycarbonyl, aryl or heteroaryl; or  $R^h$  and  $R^i$  together form  $C_{1-6}$  alkylene;  $R^j$  represents hydrogen,  $C_{1-6}$  alkyl or  $C_{1-6}$  alkoxycarbonyl; and  $R^k$  represents  $C_{1-8}$  alkyl,  $C_{1-8}$  alkoxy,  $C_{1-6}$  alkoxy( $C_{1-6}$ ) alkoxy or aryl.

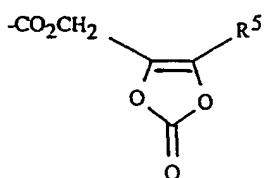
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Examples of suitable in vivo hydrolysable ester groups include, for example, acyloxyalkyl groups such as acetoxymethyl, pivaloyloxymethyl,  $\alpha$ -acetoxyethyl,  $\alpha$ -pivaloyloxyethyl, 1-(cyclohexylcarbonyloxy)prop-1-yl, and (1-aminoethyl)carbonyloxymethyl; 15 alkoxycarbonyloxyalkyl groups, such as ethoxycarbonyloxyethyl,  $\alpha$ -ethoxycarbonyloxyethyl and propoxycarbonyloxyethyl; dialkylaminoalkyl especially di-loweralkylamino alkyl groups such as dimethylaminomethyl, 20 dimethylaminoethyl, diethylaminomethyl or diethylaminoethyl; 2-(alkoxycarbonyl)-2-alkenyl groups such as 2-(isobutoxycarbonyl)pent-2-enyl and 2-(ethoxycarbonyl)but-2-enyl; lactone groups such as phthalidyl and dimethoxyphthalidyl; and esters linked to a 25 second  $\beta$ -lactam antibiotic or to a  $\beta$ -lactamase inhibitor.

A preferred in vivo hydrolysable ester group is the pivaloyloxymethyl ester.

30 A further suitable pharmaceutically acceptable in vivo hydrolysable ester group is that of the formula:

35



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wherein R<sup>5</sup> is hydrogen, C<sub>1-6</sub> alkyl or phenyl.

Suitable pharmaceutically acceptable salts of the carboxy group of the compound of formula (I) include metal salts, eg 5 aluminium, alkali metal salts such as sodium or potassium, especially sodium, alkaline earth metal salts such as calcium or magnesium, and ammonium or substituted ammonium salts, for example those with lower alkylamines such as triethylamine, hydroxy-lower alkylamines such as 10 2-hydroxyethylamine, bis-(2-hydroxyethyl)amine or tris-(2-hydroxyethyl)- amine, cycloalkylamines such as dicyclohexylamine, or with procaine, dibenzylamine, N,N-dibenzylethylene- diamine, 1-ephedamine, N-methylmorpholine, N-ethylpiperidine, 15 N-benzyl- $\beta$ -phenethylamine, dehydroabietylamine, N,N'-bisdehydro-abietylamine, ethylenediamine, or bases of the pyridine type such as pyridine, collidine or quinoline, or other amines which have been used to form salts with known penicillins and cephalosporins. Other useful salts 20 include the lithium salt and silver salt. Salts within compounds of formula (I), may be prepared by salt exchange in conventional manner.

In compounds of formula (I) or (Ia), the group X may be 25 sulphur or an oxidised sulphur atom, i.e. a sulphoxide (SO) or sulphone (SO<sub>2</sub>) group. When X is a sulphoxide group it will be understood that  $\alpha$ - and  $\beta$ -isomers may exist; both such isomers are encompassed within the scope of the present invention.

30

Examples of X include S, SO, SO<sub>2</sub> and CH<sub>2</sub>.

Preferably X is sulphur or CH<sub>2</sub>.

Advantageously, R<sup>1</sup> is hydrogen.

35

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Suitably, the cyclic ether at the 3-position of the cephalosporin nucleus is unsubstituted or substituted by up to three substituents,  $R^4$ , selected from  $C_{1-6}$  alkyl, for example methyl,  $C_{1-6}$  alkoxy, for example methoxy,  $C_{1-6}$  alkoxycarbonyl for example methoxycarbonyl,  $C_{1-6}$  alkoxy  $C_{1-6}$  alkyl, for example methoxymethyl, and  $C_{1-6}$  alkanoyloxy  $C_{1-6}$  alkyl, for example acetoxyethyl. Preferably the cyclic ether at the 3-position of the cephalosporin nucleus is unsubstituted.

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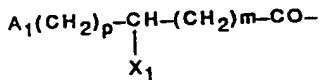
Preferably  $m$  is 1.

Preferably the cyclic ether is bonded to the cephalosporin nucleus at a ring carbon adjacent to the oxygen heteroatom.

15

Suitable acyl groups  $R^2$  include those of formulae (a) - (f):

20

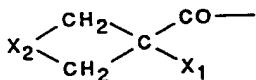


(a)

$A_3CO$  —

(b)

25

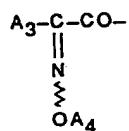


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$$A_2-X_3-(CH_2)_p-CO-$$

(d)

30



(e)

$$\begin{array}{c}
 A_3-C-CO- \\
 || \\
 A_4
 \end{array}$$

( 1 )

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wherein p is 0, 1 or 2; m is 0, 1 or 2; A<sub>1</sub> is C<sub>1-6</sub> alkyl, substituted C<sub>1-6</sub> alkyl, C<sub>3-6</sub> cycloalkyl, cyclohexenyl, cyclohexadienyl, an aromatic (including heteroaromatic) group, such as phenyl, substituted phenyl, thienyl, pyridyl, 5 or an optionally substituted thiazolyl group, a C<sub>1-6</sub> alkylthio group or C<sub>1-6</sub> alkyloxy; X<sub>1</sub> is a hydrogen or halogen atom, a carboxylic acid, carboxylic ester, sulphonic acid, azido, tetrazolyl, hydroxy, acyloxy, amino, ureido, acylamino, heterocycllamino, guanidino or acylureido group; 10 A<sub>2</sub> is an aromatic group, for example a phenyl, 2,6-dimethoxyphenyl, 2-alkoxy-1-naphthyl, 3-arylisoazolyl, or a 3-aryl-5-methylisoazolyl group, such as 3-(2-chloro-6-fluorophenyl)-5-methylisoazol-4-yl; a substituted alkyl group; or a substituted dithietane; X<sub>2</sub> 15 is a -CH<sub>2</sub>OCH<sub>2</sub>-, -CH<sub>2</sub>SCH<sub>2</sub>- or alkylene group; X<sub>3</sub> is an oxygen or sulphur atom; A<sub>3</sub> is an aryl or heteroaryl group such as phenyl, substituted phenyl, furyl, aminothiazolyl or aminothiadiazolyl in which the amino group is optionally protected; and A<sub>4</sub> is hydrogen, C<sub>1-6</sub>alkyl, C<sub>3-8</sub> cycloalkyl, 20 C<sub>3-8</sub> cycloalkyl(C<sub>1-6</sub>)alkyl, C<sub>1-6</sub> alkoxy carbonyl(C<sub>1-6</sub>) alkyl, C<sub>2-6</sub> alkenyl, carboxy(C<sub>1-6</sub>)alkyl, C<sub>2-6</sub> alkynyl, aryl or C<sub>1-6</sub>alkyl substituted by up to three aryl groups.

The term 'heteroaryl' as used herein means a heteroaromatic 25 heterocyclic ring or ring system, suitably having 5 or 6 ring atoms in each ring.

Suitably when R<sup>2</sup> is a group (a), A<sub>1</sub> is C<sub>1-6</sub> alkyl, C<sub>3-6</sub> cycloalkyl, cyclohexenyl, cyclohexadienyl, phenyl, 30 substituted phenyl such as hydroxyphenyl, thienyl or

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pyridyl; and  $X_1$  is a hydrogen or halogen atom, or a carboxy, carboxylic ester, azido, tetrazolyl, hydroxy, acyloxy, optionally protected amino, ureido, guanidino or acylureido group.

5

Suitably when  $R^2$  is a group of formula (d),  $A_2$  is phenyl,  $X_3$  is oxygen and  $p$  is 0.

Alternatively when  $R^2$  is a group of formula (e) or (f) 10 suitable values for the group  $A_3$  include those commonly found in antibacterially active cephalosporins containing a hydroxyimino, substituted hydroxyimino or vinyl group in the side chain attached to position 7 of the cephalosporin nucleus, for example phenyl, thien-2-yl, thien-3-yl, 15 fur-2-yl, fur-3-yl, pyrid-2-yl, pyrid-3-yl, pyrid-4-yl, 5-amino-1,2,4-thiadiazol-3-yl and 2-aminothiazol-4-yl in each of which the amino group is optionally protected.

Preferred groups for  $A_3$  include phenyl, 2-aminothiazol-4-yl, 20 fur-2-yl, thien-2-yl, 2-(2-chloroacetamido)thiazol-4-yl, 2-tritylamino-thiazol-4-yl, 5-amino-1,2,4-thiadiazol-3-yl and 4-aminopyrimid-2-yl.

In compounds of formula (Ia), a particularly preferred group 25 for  $A_3$  is 2-aminothiazol-4-yl.

Suitable values for the group  $A_4$  include hydrogen, methyl, ethyl, cyclopropylmethyl, triphenylmethyl (trityl), cyclobutyl, cyclopentyl, cyclohexyl, cycloheptyl, 30 cyclooctyl, phenyl, carboxymethyl, carboxypropyl and  $t$ -butoxycarbonylmethyl.

Preferred values for  $A_4$  in compounds of formula (Ia) include methyl and hydrogen.

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It will be appreciated that compounds of the invention wherein  $R^2$  is a group of formula (e) (or (f)) can exist as syn and anti (or E and Z) isomers or mixtures thereof. Both isomers are encompassed within the scope of this invention.

5

Preferably the compounds of the invention wherein  $R^2$  is a group of formula (e) have the syn configuration (i.e. have the group  $OA_4$  syn to the amide linkage) or are enriched in that isomer.

10

Similarly, when  $R^2$  is a group of formula (f), the group  $A_4$  is preferably cis to the amide linkage, i.e. when group (f) is 2-amino-thiazol-4-yl, the Z-configuration is preferred.

15 Certain compounds of the invention include an amino group which may be protected. Suitable amino protecting groups are those well known in the art which may be removed under conventional conditions without disruption of the remainder of the molecule.

20

Examples of amino protecting groups include  $C_{1-6}$  alkanoyl; benzoyl; benzyl optionally substituted in the phenyl ring by one or two substituents selected from  $C_{1-4}$  alkyl,  $C_{1-4}$  alkoxy, trifluoromethyl, halogen, or nitro;  $C_{1-4}$  25 alkoxycarbonyl; benzyloxycarbonyl or trityl substituted as for benzyl above; allyloxycarbonyl, trichloroethoxycarbonyl or chloroacetyl.

Some of the compounds of this invention may be crystallised 30 or recrystallised from solvents such as organic solvents.

In such cases solvates may be formed. This invention includes within its scope stoichiometric solvates including hydrates as well as compounds containing variable amounts of water that may be produced by processes such as 35 lyophilisation.

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Since the antibiotic compounds of the invention are intended for use in pharmaceutical compositions it will readily be understood that they are each provided in substantially pure form, for example at least 60% pure, more suitably at least 5 75% pure and preferably at least 85%, especially at least 95% pure (% are on a weight for weight basis). Impure preparations of the compounds may be used for preparing the more pure forms used in the pharmaceutical compositions; these less pure preparations of the compounds should contain 10 at least 1%, more suitably at least 5% and preferably from 10 to 49% of a compound of the formula (I) or salt thereof.

Specific compounds within this invention of formula (Ia) include the following pharmaceutically acceptable carboxylic 15 acids, salts and in-vivo hydrolysable esters:

sodium (6R, 7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(RS)-tetrahydrofuran-2-yl]-ceph-3-em-4-carboxylate;

20 pivaloyloxymethyl (6R, 7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(RS)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate;

25 sodium (6R, 7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(RS)-tetrahydropyran-2-yl]ceph-3-em-4-carboxylate;

30 pivaloyloxymethyl (6R, 7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(RS)-tetrahydropyran-2-yl]ceph-3-em-4-carboxylate;

35 (6R, 7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-hydroxyiminoacetamido]-3-[(RS)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylic acid;

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sodium (6R, 7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[ (S)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate;

5 pivaloyloxymethyl (6R, 7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[ (S)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate;

10 sodium (6R, 7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[ (R)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate;

15 pivaloyloxymethyl (6R, 7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[ (R)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate;

sodium (6R, 7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[ (RS)-tetrahydrofuran-3-yl]ceph-3-em-4-carboxylate;

20

acetoxyethyl (6R, 7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[ (S)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate;

25 sodium (6R, 7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-(5-methoxymethyltetrahydrofuran-2-yl)ceph-3-em-4-carboxylate;

30 sodium (6R, 7R)-7-[2-(2-aminothiazol-4-yl)-(Z)-pent-2-enamido]-3-[ (S)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate;

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sodium (6R, 7R)-7-[2-(2-aminothiadiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(S)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate;

5 (RS)-1-acetoxyethyl (6R, 7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(S)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate;

(6R, 7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-carboxymethoxyiminoacetamido]-3-[(RS)-tetrahydrofuran-2-yl]-ceph-3-em-4-carboxylic acid, disodium salt;

sodium (6R, 7R)-7-[(R)-2-amino-2-(4-hydroxyphenyl)acetamido]-3-[(S)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate;

15 sodium (1S, 6R, 7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(S)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate-1-oxide;

20 sodium 7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-(tetrahydrofuran-2-yl)-1-carba-1-dethiaceph-3-em-4-carboxylate;

25 sodium (6R, 7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(S)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate-1,1-dioxide;

30 (RS)-1-(propan-2-yl)oxycarbonyloxyethyl (6R, 7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(S)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate;

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sodium (6R, 7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(5R, 5R)-5-methyltetrahydrofuran-2-yl]-ceph-3-em-4-carboxylate;

5 sodium (6R, 7R)-7-[2-(furan-2-yl)-2-(Z)-methoxyiminoacetamido]-3-[(S)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate

sodium (6R, 7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(S)-5, 5-dimethyltetrahydrofuran-2-yl]-10 ceph-3-em-4-carboxylate;

sodium (6R, 7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-(5-methoxycarbonyltetrahydrofuran-2-yl)-ceph-3-em-4-carboxylate;

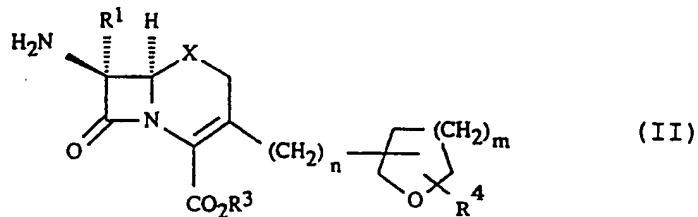
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sodium (6R, 7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[-methyltetrahydrofuran-2-yl]ceph-3-em-4-carboxylate; and

20 2-ethoxycarbonyl-(Z)-but-2-enyl (6R, 7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(S)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate.

The present invention further provides a process for the  
25 preparation of a compound of formula (I), which process  
comprises treating a compound of formula (II) or a salt  
thereof:

30



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wherein  $R^1$ ,  $CO_2R^3$ ,  $R^4$ ,  $m$ ,  $n$  and  $X$  are as hereinbefore defined, wherein any reactive groups may be protected, and wherein the amino group is optionally substituted with a group which permits acylation to take place; with an N-acylating derivative of an acid of formula (III):



10

wherein  $R^2$  is as defined with respect to formula (I) and wherein any reactive groups may be protected; and thereafter, if necessary or desired, carrying out one or more of the following steps:

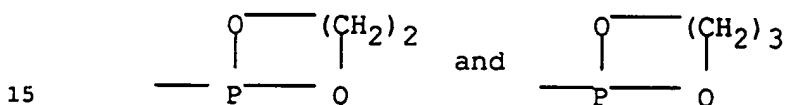
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- i) removing any protecting groups;
- ii) converting the group  $CO_2R^3$  into a different group  $CO_2R^3$ ;
- 20 iii) converting the group  $R^2$  into a different group  $R^2$ ;
- iv) converting the group  $X$  into a different group  $X$ ;
- 25 v) converting the product into a salt.

Acids of formula (III) may be prepared by methods known in the art, or methods analogous to such processes. Suitable processes include those described, for example, in UK Patent 30 2 107 307 B, UK Patent Specification No. 1,536,281, and U.K. Patent Specification No. 1,508,064.

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Suitable groups which permit acylation to take place and which are optionally present on the amino group of the starting material of the formula (II) include N-silyl, N-stannyl and N-phosphorus groups, for example trialkylsilyl groups such as trimethylsilyl, trialkyltin groups such as tri-n-butyltin, groups of formula  $-P.R^{20}R^{21}$  wherein  $R^{20}$  is an alkyl, haloalkyl, aryl, aralkyl, alkoxy, haloalkyl, aryl, aralkyl, alkoxy, haloalkoxy, aryloxy, aralkyloxy or dialkylamino group,  $R^{21}$  is the same as  $R^{20}$  or is halogen or  $R^{20}$  and  $R^{21}$  together form a ring; suitable such phosphorus groups being  $-P(OC_2H_5)_2$ ,  $-P(C_2H_5)_2$ ,



A group which may optionally be introduced onto the amino group in the compound of formula (II) is trimethylsilyl.

20 Advantageously the silylation reaction may be carried out in situ, prior to the acylation reaction, with a silylating agent that does not require concomitant addition of base. Suitable silylating agents include, for example, N-(trimethylsilyl)-acetamide,

25 N,O-bis-(trimethylsilyl)acetamide, N,O-bis(trimethylsilyl)-trifluoroacetamide, N-methyl-N-trimethylsilylacetamide, N-methyl-N-trimethylsilyl-trifluoroacetamide, N,N'-bis(trimethylsilyl)urea, and N,O-bis(trimethylsilyl)carbamate. A preferred silylating 30 agent is N,O-bis(trimethylsilyl)acetamide. The silylation reaction may suitably be carried out in an inert, anhydrous organic solvent such as dichloromethane at room temperature or at an elevated temperature, for example 30 - 60°C, preferably 40 - 50°C.

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The above process may optionally be carried out in the presence of a small quantity, for example 0.1 equivalents, of a silyl halide, for example a tri(C<sub>1-6</sub>)alkylsilyl halide, especially trimethylsilyl chloride.

5

A reactive N-acylating derivative of the acid (III) is employed in the above process. The choice of reactive derivative will of course be influenced by the chemical nature of the substituents of the acid.

10

Suitable N-acylating derivatives include an acid halide, preferably the acid chloride or bromide or alternatively a symmetrical or mixed anhydride. The acylation may be effected in the presence of an acid binding agent for 15 example, tertiary amine (such as pyridine or dimethylaniline), molecular sieves, an inorganic base (such as calcium carbonate or sodium bicarbonate) or an oxirane, which binds hydrogen halide liberated in the acylation reaction. The oxirane is preferably a (C<sub>1-6</sub>)-1,2-alkylene 20 oxide - such as ethylene oxide or propylene oxide. The acylation reaction using an acid halide may be carried out at a temperature in the range -50°C to +50°C, preferably -20°C to +20°C, in aqueous or non-aqueous media such as water, acetone, tetrahydrofuran, ethyl acetate, 25 dimethylacetamide, dimethylformamide, acetonitrile, dichloromethane, 1,2-dichloroethane, or mixtures thereof. Alternatively, the reaction may be carried out in an unstable emulsion of water-immiscible solvent, especially an aliphatic ester or ketone, such as methyl isobutyl ketone or 30 butyl acetate. The acylation with acid halide or anhydride is suitably carried out in the presence of a basic catalyst such as pyridine or 2,6-lutidine.

Acid halides may be prepared by reacting the acid (III) or a 35 salt or a reactive derivative thereof with a halogenating (eg chlorinating or brominating) agent such as phosphorus

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pentachloride, thionyl chloride, oxalyl chloride or phosgene.

Suitable mixed anhydrides are anhydrides with, for example, 5 carbonic acid monoesters, trimethyl acetic acid, thioacetic acid, diphenylacetic acid, benzoic acid, phosphorus acids (such as phosphoric, phosphorous, and phosphinic acids) or aromatic or aliphatic sulphonic acids (such as p-toluenesulphonic acid or methanesulphonic acid).

10

Alternative N-acylating derivatives of acid (III) are the acid azide, or activated esters such as esters with 2-mercaptopyridine, cyanomethanol, p-nitrophenol, 2,4-dinitrophenol, thiophenol, halophenols, including 15 pentachlorophenol, monomethoxyphenol, N-hydroxy succinimide, N-hydroxybenzotriazole, or 8-hydroxyquinoline; or amides such as N-acylsaccharins, N-acylthiazolidin-2-thione or N-acylphthalimides; or an alkylidene iminoester prepared by reaction of the acid (III) with an oxime.

20

Other reactive N-acylating derivatives of the acid (III) include the reactive intermediates formed by reaction in situ with a condensing agent such as a carbodiimide, for example, N,N'-diethyl-, dipropyl- or 25 diisopropylcarbodiimide, N,N'-di-cyclohexyl-carbodiimide, or N-ethyl-N'-[3-(dimethylamino)propyl]- carbodiimide; a suitable carbonyl compound, for example, N,N'-carbonyldiimidazole or N,N'-carbonyldi- triazole; an isoxazolinium salt, for example, 30 N-ethyl-5-phenylisoxazolinium-3-sulphonate or N-t-butyl-5-methylisoxazolinium perchlorate; or an N-alkoxycarbonyl 2-alkoxy-1,2-dihydroquinoline, such as N-ethoxycarbonyl 2-ethoxy-1,2-dihydroquinoline. Other condensing agents include Lewis acids (for example  $BBr_3 - C_6H_6$ ); 35 or a phosphoric acid condensing agent such as diethylphosphorylcyanide. The condensation reaction is

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preferably carried out in an organic reaction medium, for example, methylene chloride, dimethylformamide, acetonitrile, alcohol, benzene, dioxan or tetrahydrofuran.

5 A further method of forming the N-acylating derivative of the acid of formula (III) is to treat the acid of formula (III) with a solution or suspension preformed by addition of a carbonyl halide, preferably oxalyl chloride, or a phosphoryl halide such as phosphorus oxychloride, to a 10 halogenated hydrocarbon solvent, preferably dichloromethane, containing a lower acyl tertiary amide, preferably N,N-dimethylformamide. The N-acylating derivative of the acid of formula (III) so derived may then be caused to react with a compound of formula (II). The acylation reaction may 15 conveniently be carried out at -40° to +30°C, if desired in the presence of an acid binding agent such as pyridine. A catalyst such as 4-dimethylaminopyridine may optionally also be added. A preferred solvent for the above acylation reaction is dichloromethane.

20

The optional reduction step, the optional conversion of  $R^2$  to a different  $R^2$ ,  $CO_2R^3$  to a different  $CO_2R^3$  and  $X$  to a different  $X$ , and the optional formation of a salt, may be carried out using methods well known in the art of 25 cephalosporin and penicillin chemistry.

For example, when the group X is S, SO, or SO<sub>2</sub>, the group X may be converted into a different group X by methods of oxidation or reduction well known in the art of 30 cephalosporin and penicillin synthesis, as described, for example, in European Patent Application Publication No. 0 114 752. For example, sulphoxides (in which X is SO) may be prepared from the corresponding sulphide (in which X is S) by oxidation with a suitable oxidising agent, for example 35 an organic peracid such as m-chloroperbenzoic acid.

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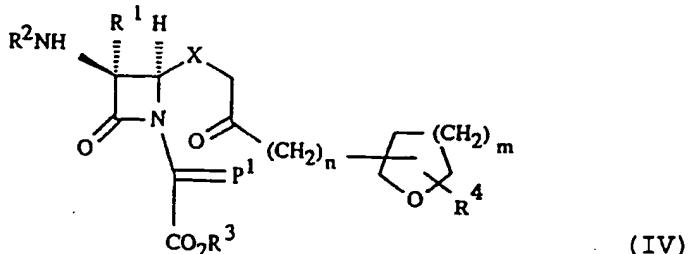
A reduction step is generally effected by processes well known in the art of  $\beta$ -lactam chemistry, for example using phosphorus trichloride in dimethylformamide.

5 In the process described hereinabove, and in the process described hereinbelow, it may be necessary to remove protecting groups. Deprotection may be carried out by any convenient method known in the art such that unwanted side reactions are minimised. Separation of unwanted by-products 10 may be carried out using standard methods.

In a further process of the invention, compounds of formula (I) may be prepared by cyclising a compound of formula (IV):

15

20



wherein X,  $R^1$ ,  $R^2$ ,  $R^4$ ,  $m$ ,  $n$  and  $CO_2R^3$  are as hereinbefore defined and  $P'$  is a phosphorus residue; and thereafter if 25 necessary or desired, carrying out one or more of the following steps:

- i) removing any protecting groups;
- 30 ii) converting the group  $CO_2R^3$  into a different group  $CO_2R^3$ ;
- iii) converting the group  $R^2$  into a different group  $R^2$ ;

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iv) converting the group X into a different group X;

v) converting the product into a salt.

5 The cyclisation reaction is an intramolecular Wittig-type reaction and is typically carried out by heating the compound of formula (IV) in an organic solvent system, for example in toluene, optionally in the presence of a suitable acid such as benzoic acid.

10

The phosphorus residue, P' is typically a trialkylphosphoranylidene residue, for example a C<sub>1-6</sub> trialkylphosphoranylidene residue such as tri-*n*-butylphosphoranylidene, or a triarylphosphoranylidene residue such as triphenylphosphoranylidene.

Where R<sup>2</sup> in a compound of formula (I) is required to be different from the group R<sup>2</sup> in the compound of formula (IV), the conversion may be effected via the intermediacy of a 20 compound of formula (II) which has an amino group at the 7-position of the cephalosporin nucleus.

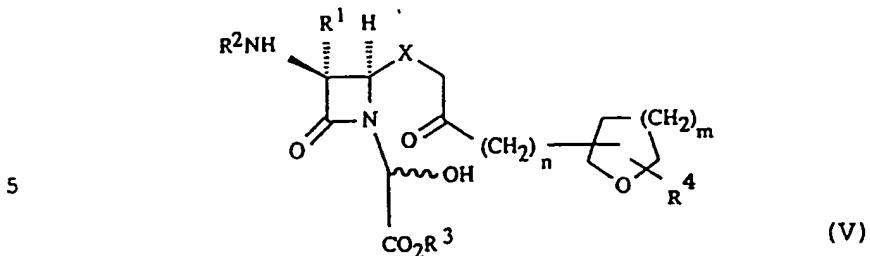
An R<sup>2</sup> side-chain may be removed by the Delft procedure commonly used in  $\beta$ -lactam chemistry. Suitable reaction 25 conditions include treatment with phosphorus pentachloride and N-methylmorpholine at reduced temperature.

Compounds of formula (II) are novel compounds and as such form part of the invention.

30

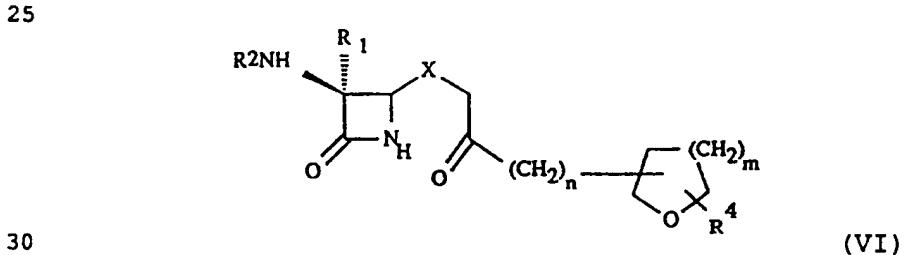
A compound of formula (IV) may be prepared from a compound of formula (V):

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wherein  $\text{X}$ ,  $\text{R}^1$ ,  $\text{R}^2$ ,  $\text{R}^4$ ,  $\text{m}$ ,  $\text{n}$  and  $\text{CO}_2\text{R}^3$  are as hereinbefore defined, by reaction with a halogenating agent, suitably a 10 chlorinating agent such as thionyl chloride, which reaction displaces the formula (V) hydroxyl group by halogen, suitably chloride, and is typically carried out at reduced temperature in an inert solvent, for example in tetrahydrofuran, in the presence of a base, typically a 15 pyridine derivative such as 2,6-lutidine. Formation of the phosphorane may be effected by treatment of the halo-intermediate with an appropriate phosphine derivative, for example tri-*n*-butylphosphine or triphenylphosphine, 20 suitably at ambient temperature in an inert solvent such as dioxan.

A compound of formula (V) may be prepared by reaction of a compound of formula (VI) :

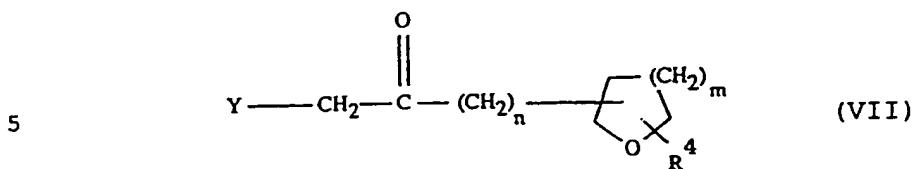


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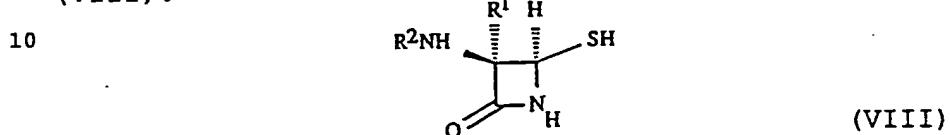
wherein  $\text{X}$ ,  $\text{R}^1$ ,  $\text{R}^2$ ,  $\text{R}^4$ ,  $\text{m}$  and  $\text{n}$  are as hereinbefore defined with an ester of glyoxylic acid ( $\text{OCHCO}_2\text{R}^3$ ) in the presence of triethylamine.

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In a typical preparation of a compound of formula (VI) in which X is sulphur, a compound of formula (VII):



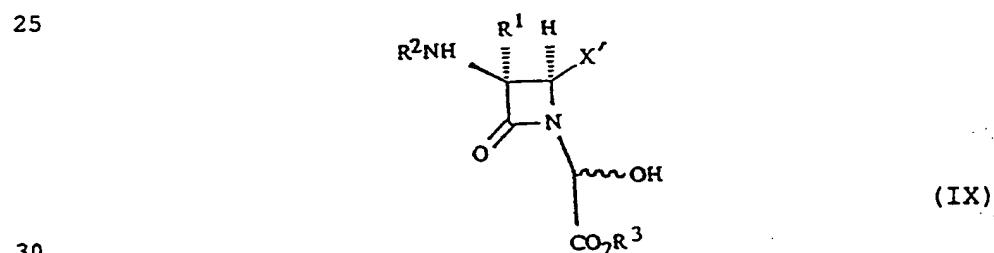
wherein Y is a leaving group and R<sup>4</sup>, m and n are as hereinbefore defined is reacted with a compound of formula (VIII):



wherein  $R^1$  and  $R^2$  are as hereinbefore defined.

15 Suitably, a leaving group Y is halogen, for example chloro. The reaction may be carried out at ambient temperature in an inert solvent, for example acetone or dimethylformamide, in the presence for a base, for example potassium carbonate.

20 A compound of formula (V) may also be prepared by reaction  
of a compound of formula (IX):



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wherein  $R^1$ ,  $R^2$  and  $CO_2R^3$  are as hereinbefore defined and  $X'$  is an  $X$ -group precursor, with a compound of formula (VII) as hereinbefore defined.

5 In a typical preparation of a compound of formula (V) in which  $X$  is sulphur, a  $Y$  leaving group in a compound of formula (VII), suitably a halogen such as chloro or bromo, is displaced by an  $X'$  mercapto group in a compound of formula (IX). The reaction may be carried out at ambient 10 temperature in an inert solvent, for example acetone, with the addition of base, for example potassium carbonate, before work-up.

Azetidin-2-one compounds of formulae (VIII) and (IX) may be 15 prepared according to known methods in heterocyclic synthetic chemistry and particularly by known methods in the art of  $\beta$ -lactam chemistry. For example a compound of formula (VIII) may be prepared according to the method of Osborne N.F. et al., J. Chem. Soc., Perkin Trans. I, 146, 20 1980.

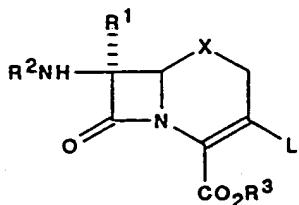
A compound of formula (IX) in which  $X'$  is a mercapto group may be prepared by ring opening of a 4-thia-2,6-diazabicyclo [3.2.0]-hept-2-ene-7-one derivative according to the method 25 of Masayuki Narisada et al., Tetrahedron Lett., 1755 (1978).

Compounds of formula (VII) are known compounds or may be prepared by standard methodology. For example, the compounds of formula (VII) in which  $Y$  is chloro or bromo may 30 be prepared from the corresponding carboxylic acid ( $Y=COOH$ ) via formation of the acid chloride followed by treatment with diazomethane and reaction of the resulting diazo compound with hydrogen chloride or hydrogen bromide.

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In a further process of the invention, compounds of formula (I) may be prepared directly by organo-cuprate displacement of a leaving group at the 3-position of a compound of formula (X):

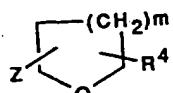
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(X)

wherein R<sup>1</sup>, R<sup>2</sup>, CO<sub>2</sub>R<sup>3</sup> and X are as hereinbefore defined and L is a leaving group, suitably a mesylate, triflate or fluorosulphonate leaving group, by reaction with a compound 15 of formula (XI):



(XI)

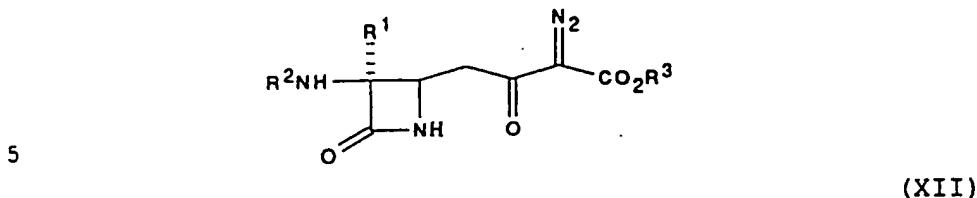
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wherein Z is an organo-cuprate group and R<sup>4</sup> and m are as hereinbefore defined.

A compound with a 3-position leaving group, L, in which X is 25 sulphur may be prepared by the procedure of Farina V. et al., J. Org. Chem., 54, 4962, (1989).

A compound with a 3-position leaving group, L, in which X is CH<sub>2</sub> may be prepared by a transition metal-catalysed 30 carbenoid insertion reaction of a diazodicarbonyl compound of formula (XII):

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wherein R<sup>1</sup>, R<sup>2</sup> and CO<sub>2</sub>R<sup>3</sup> are as hereinbefore defined, followed by reaction with an appropriate anhydride, for example triflic anhydride. Compounds of formula (XII) may be prepared by the procedure of Bodurow C. and Carr M.A.; *Tetrahedron Lett.*, 30 4801, (1989).

It should be noted that in processes of this invention Δ<sup>2</sup>-cephems may function as intermediates, in the synthetic sequences. Subsequent isomerisation steps by methods well known in cephalosporin chemistry will provide the Δ<sup>3</sup>-cephems of the invention.

20 The present invention also provides a pharmaceutical composition which comprises a compound of formula (Ia) or a pharmaceutically acceptable salt or in vivo hydrolysable ester thereof and a pharmaceutically acceptable carrier. The compositions of the invention include those in a form 25 adapted for oral, topical or parenteral use and may be used for the treatment of bacterial infection in mammals including humans.

The antibiotic compounds according to the invention may be 30 formulated for administration in any convenient way for use in human or veterinary medicine, by analogy with other antibiotics.

The composition may be formulated for administration by any 35 route, such as oral, topical or parenteral, especially oral. The compositions may be in the form of tablets, capsules,

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powders, granules, lozenges, creams or liquid preparations, such as oral or sterile parenteral solutions or suspensions.

The topical formulations of the present invention may be 5 presented as, for instance, ointments, creams or lotions, eye ointments and eye or ear drops, impregnated dressings and aerosols, and may contain appropriate conventional additives such as preservatives, solvents to assist drug penetration and emollients in ointments and creams.

10

The formulations may also contain compatible conventional carriers, such as cream or ointment bases and ethanol or oleyl alcohol for lotions. Such carriers may be present as from about 1% up to about 98% of the formulation. More 15 usually they will form up to about 80% of the formulation.

Tablets and capsules for oral administration may be in unit dose presentation form, and may contain conventional excipients such as binding agents, for example syrup, 20 acacia, gelatin, sorbitol, tragacanth, or polyvinylpyrrolidone; fillers, for example lactose, sugar, maize-starch, calcium phosphate, sorbitol or glycine; tabletting lubricants, for example magnesium stearate, talc, polyethylene glycol or silica; disintegrants, for example 25 potato starch; or acceptable wetting agents such as sodium lauryl sulphate. The tablets may be coated according to methods well known in normal pharmaceutical practice. Oral liquid preparations may be in the form of, for example, aqueous or oily suspensions, solutions, emulsions, syrups or 30 elixirs, or may be presented as a dry product for reconstitution with water or other suitable vehicle before use. Such liquid preparations may contain conventional additives, such as suspending agents, for example sorbitol, methyl cellulose, glucose syrup, gelatin, hydroxyethyl 35 cellulose, carboxymethyl cellulose, aluminium stearate gel

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or hydrogenated edible fats, emulsifying agents, for example lecithin, sorbitan monooleate, or acacia; non-aqueous vehicles (which may include edible oils), for example almond oil, oily esters such as glycerine, propylene glycol, -or 5 ethyl alcohol; preservatives, for example methyl or propyl p-hydroxybenzoate or sorbic acid, and, if desired, conventional flavouring or colouring agents.

Suppositories will contain conventional suppository bases, 10 e.g. cocoa-butter or other glyceride.

For parenteral administration, fluid unit dosage forms are prepared utilizing the compound and a sterile vehicle, water being preferred. The compound, depending on the vehicle and 15 concentration used, can be either suspended or dissolved in the vehicle. In preparing solutions the compound can be dissolved in water for injection and filter sterilised before filling into a suitable vial or ampoule and sealing.

20 Advantageously, agents such as a local anaesthetic, preservative and buffering agents can be dissolved in the vehicle. To enhance the stability, the composition can be frozen after filling into the vial and the water removed under vacuum. The dry lyophilized powder is then sealed in 25 the vial and an accompanying vial of water for injection may be supplied to reconstitute the liquid prior to use.

Parenteral suspensions are prepared in substantially the same manner except that the compound is suspended in the vehicle instead of being dissolved and sterilization cannot 30 be accomplished by filtration. The compound can be sterilised by exposure to ethylene oxide before suspending in the sterile vehicle. Advantageously, a surfactant or wetting agent is included in the composition to facilitate uniform distribution of the compound.

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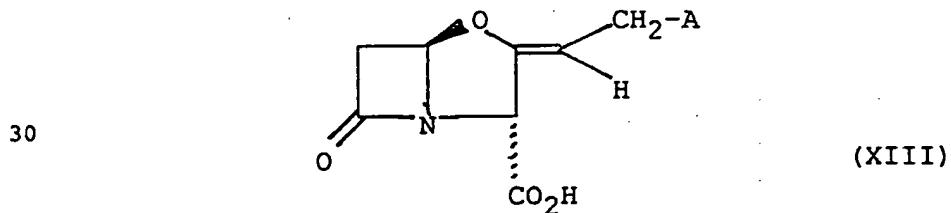
The compositions may contain from 0.1% by weight, preferably from 10-60% by weight, of the active material, depending on the method of administration. Where the compositions 5 comprise dosage units, each unit will preferably contain from 50-500 mg of the active ingredient. The dosage as employed for adult human treatment will preferably range from 100 to 3000 mg per day, for instance 1500 mg per day depending on the route and frequency of administration.

10 Such a dosage corresponds to 1.5 to 50 mg/kg per day. Suitably the dosage is from 5 to 20 mg/kg per day.

No unacceptable toxicological effects are expected when a compound of formula (Ia) or a pharmaceutically acceptable 15 salt or in vivo hydrolysable ester thereof is administered in the above-mentioned dosage range.

The compound of formula (Ia) may be the sole therapeutic agent in the compositions of the invention or a combination 20 with other antibiotics or with a  $\beta$ -lactamase inhibitor may be employed.

Advantageously, the compositions also comprise a compound of formula (XIII) or a pharmaceutically acceptable salt or 25 ester thereof:

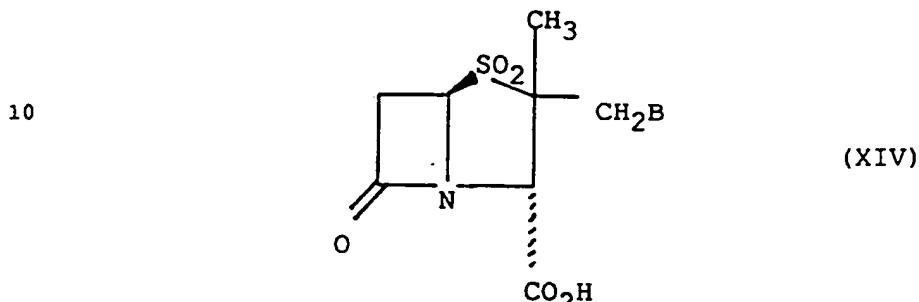


wherein

35 A is hydroxyl, substituted hydroxyl, thiol, substituted thiol, amino, mono- or di-hydrocarbyl- substituted amino, or mono- or di-acylamino; an optionally substituted triazolyl group; or an optionally substituted tetrazolyl group as described in EP-A-0 053 893.

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A further advantageous composition comprises a compound of formula (Ia) or a pharmaceutically acceptable salt or in vivo hydrolysable ester thereof together with a compound of formula (XIV) or a pharmaceutically acceptable salt or in vivo hydrolysable ester thereof:



15

wherein

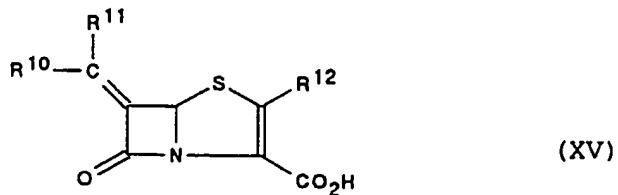
B represents hydrogen, halogen or a group of formula:



in which R<sup>8</sup> and R<sup>9</sup> are the same or different and each represents hydrogen, C<sub>1-6</sub> alkoxy carbonyl or carboxy, or a pharmaceutically acceptable salt thereof.

Further suitable  $\beta$ -lactamase inhibitors include 6-alkylidene penems of formula (XV):

30



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or a pharmaceutically acceptable salt or in vivo hydrolysable ester thereof, wherein R<sup>10</sup> and R<sup>11</sup> are the same or different and each represents hydrogen, or a C<sub>1</sub>-10-5 hydrocarbon or heterocyclic group optionally substituted with a functional group; and R<sup>12</sup> represents hydrogen or a group of formula R<sup>13</sup> or -SR<sup>13</sup> where R<sup>13</sup> is an optionally substituted C<sub>1</sub>-10 hydrocarbon or heterocyclic group, as described in EP-A-0 041 768.

10

Further suitable  $\beta$ -lactamase inhibitors include 6 $\beta$ -bromopenicillanic acid and pharmaceutically acceptable salts and in vivo hydrolysable esters thereof and 6 $\beta$ -iodopenicillanic acid and pharmaceutically acceptable 15 salts and in vivo hydrolysable esters thereof described in, for example, EP-A-0 410 768 and EP-A-0 154 132 (both Beecham Group).

Such compositions of this invention which include a 20  $\beta$ -lactamase inhibitory amount of a  $\beta$ -lactamase inhibitor are formulated in a conventional manner using techniques and procedures per se known in the art.

The antibiotic compounds of the present invention are active 25 against a wide range of organisms including both Gram-negative organisms such as E.coli and Gram-positive organisms such as S.aureus.

The following Examples illustrate the preparation of 30 compounds of the invention and intermediates thereto. The following biological data illustrate the activity of compounds of the invention in the form of MIC values (minimum inhibitory concentration) against a sample E.coli organism (NCTC 10418) and a sample S.aureus organism 35 (S.aureus Oxford).

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Example 1

Sodium (6R, 7R)-7-[2-(2-Aminothiazol-4-yl)-2-(2-methoxy-iminoacetamido]-3-[(RS)-tetrahydrofuran-2-yl]ceph-3-em-4-5 carboxylate

(a) (RS)-2-Chloroacetyl tetrahydrofuran

Oxalyl chloride (5.2ml, 60mmol) and DMF (1 drop) were added 10 to (RS)-2-tetrahydrofuroic acid (W.E. Kaufmann and R. Adams, J.Amer.Chem.Soc., 1923, 45, 3029) (4.64g, 40mmol) in dichloromethane (25ml). The mixture was stirred 1h, evaporated in vacuo, dichloromethane added and reevaporated to give 2-tetrahydrofuroyl chloride,  $\nu_{\text{max}}(\text{CH}_2\text{Cl}_2)$  1795cm<sup>-1</sup>. 15 2-Tetrahydrofuroyl chloride in ether (25ml) and dichloromethane (10ml) was added dropwise to an ice bath cooled solution of diazomethane (ca 80mmol) in ether (150ml). The reaction mixture was stirred 0.25h then a stream of hydrogen chloride gas passed into the solution for 20 ca 2 minutes then stirred a further 0.25h, washed with saturated brine, dried, concentrated and flash chromatographed on silica gel eluting with 5, 7.5 and 10% ethyl acetate in hexane to provide the title compound (2.46g, 41%); (Found:  $\text{M}^+$ , 148.0279.  $\text{C}_6\text{H}_9\text{ClO}_2$  requires  $\text{M}$ , 25 148.0291);  $\nu_{\text{max}}(\text{CH}_2\text{Cl}_2)$  1739, 1395, 1071 and 936cm<sup>-1</sup>;  $\delta_{\text{H}}(\text{CDCl}_3, 250\text{MHz})$  1.8-2.4 (4H, m) and 3.9-4.6 (5H, m).

(b) (3R, 4R)-3-Phenoxyacetamido-4-[(RS)-tetrahydrofuran-2-ylcarbonylmethylthio]azetidin-2-one

30 (RS)-2-Chloroacetyl tetrahydrofuran (2.46g, 16.5mmol), (3R, 4R)-4-mercaptop-3-phenoxyacetamidoazetidin-2-one (4.157g, 16.5mmol) and potassium carbonate (2.227g 16.5mmol) in DMF (10ml) were stirred for 2h, diluted with ethyl acetate, 35 washed twice with water and with brine, dried concentrated

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and flash chromatographed eluting with 40, 30, 20, 10 and 0% hexane in ethyl acetate to give the title compound as a foam (3.547g, 59%);  $\nu_{\text{max}}$  ( $\text{CH}_2\text{Cl}_2$ ) 3405, 1785, 1693, 1520, 1496 and 1240  $\text{cm}^{-1}$ ;  $\delta$  ( $\text{CDCl}_3$ , 250MHz) 1.9-2.3 (4H, m), 3.42 and 3.62, 5 3.46 and 3.56 (together 2H, 2 ABq,  $J$  15.8Hz, 15.4Hz), 3.85-4.0 (2H, m), 4.4-4.5 (1H, m), 4.58 (2H, s), 5.01, 5.04 (together 1H, 2d,  $J$  4.7Hz), 5.59 (1H, dd,  $J$  8.8, 4.5Hz) 6.62, 6.68 (together 1H, 2s), 6.9-7.4 (5H, m) and 7.45, 7.47 (together 1H, 2d,  $J$  8.8Hz). [Mass spectrum:  $\text{M}^+$  (364)].

10

(c) t-Butyl (RS)-2-Hydroxy-2-[(3R,4R)-3-phenoxyacetamido-4-[(RS)-tetrahydrofuran-2-ylcarbonylmethylthio]azetidin-2-on-1-yl]acetate

15 0.5M t-Butyl glyoxylate in 1,2-dichloroethane (20ml) and triethylamine (140  $\mu$ l, 1mmol) were added to (3R,4R)-phenoxyacetamido-4-[(RS)-tetrahydrofuran-2-ylcarbonylmethylthio]azetidin-2-one (3.547g, 9.7mmol) in 1,2-dichloroethane (10ml). The mixture was stirred 1h, 20 concentrated in vacuo and flash chromatographed eluting with 50, 60, 70% ethyl acetate in hexane (3.663g, 76%);  $\nu_{\text{max}}$  ( $\text{CH}_2\text{Cl}_2$ ) 3471, 3407, 1782, 1736, 1692, 1521, 1290, 1154 and 1083  $\text{cm}^{-1}$ ;  $\delta_{\text{H}}$  ( $\text{CDCl}_3$ , 250MHz) 1.53 (9H, s), 1.85-2.25 (4H, m), 3.4-3.7 (2H, m), 3.8-4.0 (2H, m), 4.3-4.45 (1H, m), 4.57 m, 5.07, 5.09, 5.16, 5.18 (together 1H, 4d,  $J$  4.8Hz), 25 5.25-5.45 (1H, m), 5.48, 5.58 (together 1H, 2dd,  $J$  4.8, 8.8Hz), 6.9-7.4 (5H, m) and 7.41, 7.56 (together 1H, 2d,  $J$  8.7Hz). [Mass spectrum: +ve ion (thioglycerol)  $\text{MH}^+$  (495)].

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(d) t-Butyl 2-[(3R,4R)-3-Phenoxyacetamido-4-[(RS)-tetrahydrofuran-2-ylcarbonylmethylthio]azetidin-2-on-1-yl]-2-tri-n-butylphosphoranylideneacetate

5 Thionyl chloride (0.81ml, 11.1mmol) in THF (5ml) was added dropwise to the hydroxy compound (3.663g, 7.4mmol) and 2,6-lutidine (1.29ml, 11.1mmol) in THF (15ml) at -20°C. The mixture was stirred 0.5h, filtered and the filtrate evaporated in vacuo, toluene added and re-evaporated to give 10 t-butyl (RS)-2-chloro-2-[(3R,4R)-3-phenoxyacetamido-4-[(RS)-tetrahydrofuran-2-ylcarbonylmethylthio]azetidin-2-on-1-yl]acetate as a foam (4.222g).

To the crude chloro compound in dioxan (10ml) was added 15 tri-n-butylphosphine (4.06ml, 16.3mmol), the solution stirred 0.75h, [Bdiluted with ethyl acetate, washed with dilute sodium hydrogen carbonate solution, water and brine, dried concentrated and flash chromatographed on silica gel eluting with 30, 40, 50, 60, 70, 80% ethyl acetate in hexane to 20 give the title compound as a foam (3.827g, 76%);  $\nu_{\text{max}}(\text{CH}_2\text{Cl}_2)$  3417, 1764, 1731, 1690, 1628, 1523, 1171 and 1082cm<sup>-1</sup> [Mass spectrum: +ve ion (thioglycerol)  $\text{MH}^+$  (679)].

(e) t-Butyl (6R,7R)-7-Phenoxyacetamido-3-[(RS)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate

The phosphorane (3.827g) and benzoic acid (20mg) in toluene (75ml) were purged with argon then heated under argon in an oil bath at 130°C for 6h. The solution was left to cool and 30 flash chromatographed on silica gel eluting with 30% ethyl acetate in hexane to give the title compound as a foam (2.267g, 87%);  $\nu_{\text{max}}(\text{CH}_2\text{Cl}_2)$  3406, 1785, 1697, 1519, 1155 and 1054cm<sup>-1</sup>;  $\delta_{\text{H}}(\text{CDCl}_3, 250\text{MHz})$  1.53, 1.54 (together 9H, 2s), 1.5-2.5 (4H, m), 3.29 and 3.61, 3.39 and 3.56 (together 2H,

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2ABq, J18.6, 18.0Hz), 3.8-4.0 (2H, m), 4.57 (2H, s), 4.9-5.0, 5.05-5.2 (together 1H, 2m), 5.01, 5.02 (together 1H, 2d, J4.8Hz), 5.84, 5.91 (together 1H, 2dd, J4.8, 9.4Hz) and 6.9-7.4 (6H, m). [Mass spectrum +ve ion (3-nitrobenzyl 5 alcohol, sodium acetate)  $MNa^+$ (483)].

(f) t-Butyl (6R,7R)-7-Amino-3-(tetrahydrofuran-2-yl)ceph-3-em-4-carboxylate

10 Phosphorus pentachloride (1.538g, 7.5mmol) in dichloromethane (39ml) was added to t-butyl(6R,7R)-7-phenoxyacetamido-3-[(RS)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate (2.267g, 4.9mmol) and N-methylmorpholine (1.1ml, 10mmol) in dichloromethane (20ml) at -25°C. The 15 reaction was stirred at -10±5°C for 0.75h then methanol (10ml) added all at once, stirred 0.75h then water (20ml) added and stirred vigorously for 1h. The dichloromethane was evaporated in vacuo, the aqueous residue washed with ether then adjusted to pH7 with ammonium hydroxide in the 20 presence of ethyl acetate. The mixture was extracted twice with ethyl acetate, the extracts dried, concentrated and flash chromatographed on silica gel eluting with 30,40,50% ethyl acetate in hexane to give the more mobile (S)-diastereoisomer of the title compound (0.431g, 27%); (Found: 25  $M^+$ , 326.1299.  $C_{15}H_{22}N_2O_4S$  requires  $M$ , 326.1300);  $\nu_{max}$  ( $CH_2Cl_2$ ) 1777, 1716, 1158 and 1052cm<sup>-1</sup>;  $\delta_H$  ( $CDCl_3$ , 250MHz) 1.52 (9H, s), 1.55-1.8 (1H, m), 1.85-2.05 (4H, m), 2.3-2.45 (1H, m), 3.30 and 3.59 (2H, ABq, J18.4Hz), 3.8-4.0 (2H, m), 4.75 (1H, d, J5.0Hz) and 4.9-5.0 (2H, m). 30 Further elution with 60% ethyl acetate in hexane gave the more polar (R)-diastereoisomer (0.533g, 33%); (Found:  $M^+$  326.1299.  $C_{15}H_{22}N_2O_4S$  requires  $M$ , 326.1300)  $\nu_{max}$  ( $CH_2Cl_2$ ) 1776, 1721, 1158 and 1052cm<sup>-1</sup>;  $\delta_H$  ( $CDCl_3$ , 250MHz) 1.41 (2H, bs), 1.54 (9H, s), 1.6-1.85 (1H, m), 1.9-2.05 (2H, m), 2.05-2.2 (1H, m), 3.40 and 3.55 (2H, ABq, J17.8Hz) 3.8-4.0.

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(2H, m), 4.67 (1H, d, J5.0Hz), 4.93 (1H, d, J5.0Hz),  
5.0-5.15 (1H, m).

(g) t-Butyl (6R,7R)-7-[2-(Z)-Methoxyimino-2-(2-trityl-5-aminothiazol-4-yl)acetamido]-3-(tetrahydrofuran-2-yl)-ceph-3-em-4-carboxylate

Mesyl chloride (141 $\mu$ l, 1.8mmol) was added to 2-(Z)-methoxyimino-2-(2-tritylaminothiazol-4-yl)acetic acid 10 hydrochloride (0.744g, 1.65mmol) and N,N-diisopropylethylamine (576 $\mu$ l, 3.3mmol) in DMF (5ml) at -40°C. The reaction mixture was stirred 0.5h at -30±10°C then t-butyl (6R,7R)-7-amino-3-(tetrahydrofuran-2-yl)ceph-3-em-4-carboxylate, more mobile diastereoisomer (0.431g, 15 1.3mmol) in DMF (5ml) followed by pyridine (147 $\mu$ l, 1.8mmol) were added. Stirred 1h without further cooling then diluted with ethyl acetate, washed twice with water and with brine, dried, concentrated and flash chromatographed on silica gel eluting with 30,35 and 40% ethyl acetate in hexane to give 20 the title compound as a foam (0.83g, 84%);  $\nu_{\text{max}}$  (CH<sub>2</sub>Cl<sub>2</sub>) 3396, 3277, 1782, 1732, 1683, 1526, 1248, 1156 and 1051cm<sup>-1</sup>;  $\delta_{\text{H}}$  [(CD<sub>3</sub>)<sub>2</sub>SO, 250MHz] 1.47 (9H, s), 1.55-1.75 (1H, m), 1.8-2.0 (2H, m), 2.05-2.2 (1H, m), 3.44 and 3.50 (2H, ABq, J18.3Hz), 3.65-3.95 (2H, m), 3.81 (3H, s), 4.6-4.7 (1H, m), 25 5.14 (1H, d, J4.8Hz), 5.66 (1H, dd, J4.8, 7.9Hz), 6.70 (1H, s), 7.2-7.4 (15H, m), 8.88 (1H, s) and 9.54 (1H, d, J7.9Hz). [Mass spectrum: +ve ion (3-nitrobenzyl alcohol, sodium acetate) MNa<sup>+</sup> (774)].

(h) Sodium (6R-7R)-7-[2-(2-Aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(RS)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate

t-Butyl (6R,7R)-7-[2-(Z)-methoxyimino-2-(2-tritylaminothiazol-4-yl)acetamido]-3-(tetrahydrofuran-2-yl)ceph-3-em-4-carboxylate, single diastereoisomer (0.832g, 1.1mol) in 0.1M

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hydrochloric acid in 90% formic acid (11ml) was stood for 1h, concentrated hydrochloric acid (200 $\mu$ l) added and left for a further 1.5h then evaporated to dryness in vacuo. The residue in water (ca 5ml) was adjusted to pH6.5 with 1M sodium hydroxide solution and chromatographed on HP20SS eluting with 0,1,2 and 3% THF in water. Fractions containing the product, h.p.l.c. analysis, were combined, concentrated and freeze dried to give the title compound as a mixture of diastereoisomers (271mg, 52%);  $\nu_{\text{max}}$  (KBr) 1762, 10 1669, 1603, 1530, 1388 and 1039cm $^{-1}$ ;  $\delta_{\text{H}}$  [(CD<sub>3</sub>)<sub>2</sub>SO, 250MHz] 1.4-2.05 (4H, m), 3.19 and 3.36, 3.26 and 3.83 (together 2H, 2ABq, J17.5, 16.8Hz), 3.55-3.85 (1H, m), 3.83 (3H, s), 4.85-4.95, 5.15-5.25 (together 2H, 2m), 4.96, 4.97 (together 1H, 2d, J4.7Hz), 5.49, 5.53 (together 1H, 2dd, J4.7, 7.9Hz), 15 6.74, 6.75 (together 1H, 2s), 7.24 (2H, s), 9.49, 9.52 (together 1H, 2d, J7.9Hz) [Mass spectrum +ve ion (thioglycerol)  $\underline{\text{M}}\text{H}^+$  (476),  $\underline{\text{M}}\text{Na}^+$  (498)].

The same mixture of diastereoisomers was obtained by 20 progressing the other diastereoisomer isolated in stage (f).

#### Example 2

Pivaloyloxymethyl (6R,7R)-7-[2-(2-Aminothiazol-4-yl)-2-(Z)-  
25 methoxyiminoacetamido]-3-[(RS)-tetrahydrofuran-2-yl]ceph-3-  
em-4-carboxylate

Pivaloyloxymethyl bromide (0.15g) and sodium iodide (0.15g) in acetone (1ml) were stirred 0.5h, filtered and the 30 filtrate evaporated to give the iodide. This in toluene (0.5ml) was added to sodium (6R,7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(RS)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate (0.191g) in N-methyl-

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pyrrolidone (1ml) and stirred 0.5h. The reaction mixture was diluted with ethyl acetate, washed twice with water and with brine, dried, concentrated and flash chromatographed on silica gel eluting with 80% ethyl acetate in hexane to give 5 the title compound (130mg, 57%);  $\nu_{\text{max}}$  (CH<sub>2</sub>Cl<sub>2</sub>) 3478, 3391, 1787, 1752, 1685, 1125, 1098 and 1052cm<sup>-1</sup>;  $\delta_{\text{H}}$  (CDCl<sub>3</sub>, 250MHz), 1.23 (9H, s), 1.6-2.5 (4H, m), 3.37 and 3.66, 3.43 and 3.62 (together 2H, 2ABq, J18.8, 17.8Hz), 3.8-4.05 (2H, m), 4.10 (3H, s), 4.85-5.0, 5.15-5.25 (together 1H, 2m), 10 5.07, 5.08 (together 1H, 2d, J4.8, 4.7Hz), 5.8-6.05 (3H, m), 6.95, 6.96 (together 1H, 2s) and 7.54, 7.65 (together 1H, 2d, J8.8, 8.5Hz). [Mass spectrum: +ve ion (thioglycerol)  $\text{M}^+$  568)].

15 Example 3

Sodium (6R,7R)-7-[2-(2-Aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(RS)-tetrahydropyran-2-yl]ceph-3-em-4-carboxylate

20

(a) Tetrahydropyran-2-carboxylic acid

3,4-Dihydro-2H-pyran-2-carboxylic acid, sodium salt (5.0g) in water (30ml) was treated with 10% Palladium on carbon 25 catalyst (0.2g) and the mixture hydrogenated until there was no further uptake of hydrogen. The mixture was filtered through Kieselguhr, the filtrate passed through a column of 'Amberlite IR120(H<sup>+</sup>)', evaporated in vacuo and the residue dissolved in dichloromethane, dried and evaporated to give 30 the title compound as colourless oil. (3.3g, 76%); (Found:  $\text{M}^+$ , 130.0631. C<sub>6</sub>H<sub>10</sub>O<sub>3</sub> requires M, 130.0630;  $\nu_{\text{max}}$  (CH<sub>2</sub>Cl) 3500-2750 (v.br), 1772, 1725cm<sup>-1</sup>;  $\delta_{\text{H}}$  (CDCl<sub>3</sub>), 1.5-1.7 (4H, m), 1.8-2.1 (2H, m), 3.50-3.59 (1H, m), 3.99-4.14 (2H, m) and 7.28 (1H, br.s).

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(b) 2-(2-Chloroacetyl)tetrahydropyran

Tetrahydropyran-2-carboxylic acid (3.3g) in dry dichloromethane (60ml) was treated with oxalyl chloride 5 (4.8g, 3.3ml) and DMF (2-3 drops). After the initial effervescence had ceased the mixture was left for a further 1h at ambient temperature. The solvent and excess oxalyl chloride were removed in vacuo and the resultant oil [ $\nu_{\text{max}}$  (CH<sub>2</sub>Cl<sub>2</sub>) 1830cm<sup>-1</sup>] was dissolved in dichloromethane 10 (20ml). This acid chloride solution was then added dropwise to a freshly prepared ethereal solution of diazomethane (ca 2 fold excess) cooled to 0-5°C, t.l.c. analysis (60% ethyl acetate in hexane) showed a single mobile spot, i.r. spectrum of a sample showed clean conversion to the 15 diazoketone [ $\nu_{\text{max}}$  (CH<sub>2</sub>Cl<sub>2</sub>) 2100cm<sup>-1</sup>]. Hydrogen chloride gas was bubbled through the solution until no further starting material was observed by t.l.c. The mixture was washed with brine, dried and the solvent removed in vacuo and the residue purified by flash chromatography on silica gel. The 20 title compound was obtained as a pale yellow oil, (2.8g, 68%);  $\nu_{\text{max}}$  (CH<sub>2</sub>Cl<sub>2</sub>) 1740cm<sup>-1</sup>;  $\delta_{\text{H}}$  (CDCl<sub>3</sub>) 1.4-1.7 (4H, m), 1.91-1.98 (2H, m), 3.42-3.53 (1H, m), 3.95-4.07 (2H, m) and 4.48 (2H, s) [Mass spectrum: +ve ion (NH<sub>3</sub>), MH<sup>+</sup> (163), MNH<sub>4</sub><sup>+</sup> (180)].

25

(c) (3R,4R)-3-Phenylacetamido-4-[(RS)-tetrahydro-  
pyran-2-ylcarbonylmethylthio]azetidin-2-one

3R, 4R-Mercapto-3-phenylacetamidoazetidin-2-one (2.6g) and 30 2-(2-chloroacetyl)tetrahydropyran (1.6g) in DMF (20ml) were treated with potassium carbonate (1.6g) at ambient temperature for ca 2h until t.l.c. (80% ethylacetate in hexane) showed loss of starting material. The reaction mixture was diluted with ethyl acetate, washed with water 35 (x3), brine, dried and concentrated. The title compound was obtained by flash chromatography (60%, 70% ethyl acetate in hexane, ethyl acetate) as a mixture of diastereoisomers as a

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colourless foam (1.7g, 70%);  $\nu_{\text{max}}$  ( $\text{CH}_2\text{Cl}_2$ ), 3380 (w), 1783, 1726, 1684  $\text{cm}^{-1}$ ;  $\delta_{\text{H}}$  ( $\text{CDCl}_3$ ) 1.3-1.7 (4H, m), 1.8-2.0 (2H, m), 3.3-3.6 (3H, m), 3.66 (2H, s), 3.86-3.90 (1H, m), 4.03-4.07 (1H, m), 4.92 (1H, d,  $J$ 4.6Hz), 5.51 (1H, dd,  $J$ 4.4, -8.6Hz) 5 6.42 (d,  $J$ 8.7Hz), 6.48, 6.51 (together 1H, 2s) and 7.27-7.36 (5H, m). [Mass spectrum:  $M^+$  (362)].

(d) t-Butyl (RS)-2-Hydroxy-2-[(3R,4R)-3-phenylacetamido-4-[(RS)-tetrahydropyran-2-ylcarbonylmethylthio]azetidin-2-on-1-yl]acetate

(3R, 4R)-3-Phenylacetamido-4-[(RS)-tetrahydropyran-2-ylcarbonylmethylthio]azetidin-2-one (1.7g) in 1,2-dichloroethane (20ml) was successively treated with 0.5M 15 t-butyl glyoxylate in 1,2-dichlorethane (10ml) and triethylamine (50mg, 70 $\mu$ l) and monitored by t.l.c. (ethyl acetate) until no starting material remained. The reaction mixture was concentrated and flash chromatography (70% ethyl acetate in hexane, ethyl acetate) to afford the title 20 compound as a yellow foam (1.9g, 82%);  $\nu_{\text{max}}$  ( $\text{CH}_2\text{Cl}_3$ ) 3400 (w), 1780, 1736, 1687  $\text{cm}^{-1}$ ;  $\delta_{\text{H}}$  ( $\text{CDCl}_3$ ) 1.49 (9H, s) overlapping 1.44-1.61 (4H, m), 1.8-2.0 (2H, m), 3.35-3.58 (3H, m), 3.65 (2H, s), 3.81-3.92 (1H, m), 4.01-4.06 (1H, m), 4.28-4.43 (1H, m), 4.99, 5.00, 5.07 (together 1H, 3d, 25  $J$ 4.7Hz), 5.21, 5.32, 5.33 (together 1H, 3d,  $J$ 6.8, 7.7, 7.6Hz), 5.42, 5.50 (together 1H, 2dd,  $J$ 4.8, 8.7Hz, 6.35, 6.36, 6.61 (together 1H, 3d,  $J$ 8.7Hz) and 7.27-7.38 (5H, m).

(e) t-Butyl 2-[(3R,4R)-3-Phenylacetamido-4-[(RS)-tetrahydropyran-2-ylcarbonylmethylthio]azetidin-2-on-1-yl]-2-tri-n-butylphosphoranylideneacetate

t-Butyl 2-hydroxy-2-[(3R,4R)-3-phenylacetamido-4-[(RS)-

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tetrahydropyran-2-ylcarbonylmethylthio]azetidin-2-on-1-yl]acetate (1.9g) in dry THF (10ml) was treated with 2,6-lutidine (0.62g, 0.67ml) followed by thionyl chloride (0.69g, 0.42ml) in THF (5ml) dropwise at <-20°C under argon. The reaction mixture was allowed to warm slowly to ca 0°C at which point no starting material was observed by t.l.c. (ethyl acetate). The reaction mixture was filtered and solvent removed in vacuo, the residue dissolved in toluene and evaporated to afford crude *t*-butyl (RS)-2-chloro-2-[(3*R*,4*R*)-3-phenylacetamido-4-[(RS)-tetrahydropyran-2-ylcarbonylmethylthio]azetidin-2-on-1-yl]acetate as a brown gum. This was dissolved in dry dioxan (10ml) and treated with tri-*n*-butylphosphine (1.79g, 2.2ml). The reaction mixture was stirred until loss of starting material was observed by t.l.c. (ethyl acetate) ca 0.5h. After removal of solvent in vacuo the title compound was obtained by flash chromatography (eluting with 50, 60, 80% ethyl acetate in hexane, ethyl acetate) as a pale brown foam (1.95g, 75%);  $\nu_{\text{max}}$  (CH<sub>2</sub>Cl<sub>2</sub>) 3417(w), 1762, 1681, 1625cm<sup>-1</sup>. [Mass spectrum +ve ion (thioglycerol) MH<sup>+</sup> (677)].

(f) *t*-Butyl (6*R*,7*R*)-7-Phenylacetamido-3-[(RS)-tetrahydropyran-2-yl]ceph-3-em-4-carboxylate

*t*-Butyl 2-[(3*R*,4*R*)-3-phenylacetamido-4-[(RS)-tetrahydropyran-2-yl]carbonylmethylthio]azetidin-2-on-1-yl]-2-tri-*n*-butylphosphoranylideneacetate (1.95g) in dry toluene (50ml) was refluxed for 8h under argon. The solvent was removed in vacuo and the title compound obtained by flash chromatography (30% ethyl acetate in dichloromethane) as a yellow foam (1.15g, 87%);  $\nu_{\text{max}}$  (CH<sub>2</sub>Cl<sub>2</sub>) 3415(w), 1783, 1721, 1687cm<sup>-1</sup>;  $\delta_{\text{H}}$  (CDCl<sub>3</sub>), 1.54, 1.56 (together 9H, 2s) overlapping 1.46-1.68 (4H, m), 1.76-1.94 (2H, m), 3.42-3.68 (5H, m), 3.97-4.06 (2H, m), 4.52-4.65 (1H, m), 4.98 (1H, d, J4.8Hz), 5.68, 5.71 (together 1H, dd, J4.7) and 7.23-7.36

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(5H, m). [Mass spectrum: +ve ion (3-nitrobenzyl alcohol, sodium acetate)  $\underline{MNa}^+$  (481)].

(g) t-Butyl (6R,7R)-7-Amino-3-[(RS)-tetrahydropyran-5-2-yl]ceph-3-em-4-carboxylate

t-Butyl (6R,7R)-7-phenylacetamido-3-[(RS)-tetrahydro-pyran-2-yl]ceph-3-em-4-carboxylate (1.1g) in dry dichloromethane (50ml) at  $-20^{\circ}\text{C}$  under argon was successively 10 treated with N-methylmorpholine (0.55g, 0.6ml) and phosphorus pentachloride (0.65g as 16.25ml of a 40mg/ml solution in dry dichloromethane) and stirred at  $-20^{\circ}\text{C}$  for 0.75h. Methanol (50ml) was added and reaction mixture allowed to warm to ambient temperature over a period of ca 15 0.5h. Water (50ml) was added and reaction mixture stirred vigourously for a further 0.5h. The dichloromethane was removed in vacuo, ethyl acetate added and aqueous layer adjusted to pH8 with 0.880 ammonia and reextracted with ethyl acetate. The organic extracts were washed with water, 20 brine, dried, concentrated and flash chromatographed on silica gel (eluting with 70,80% ethyl acetate in hexane, ethyl acetate). The first isomer to be eluted (isomer A) was obtained as a white foam (300mg, 37%);  $\nu_{\text{max}}$  ( $\text{CH}_2\text{Cl}_2$ ) 1776, 1717 $\text{cm}^{-1}$ ;  $\delta_{\text{H}}$  ( $\text{CDCl}_3$ ), 1.53 (9H, s) overlapping 1.4-1.7 25 (4H, m), 1.73-1.97 (2H, m), 3.35-3.55 (1H, m) overlapping 3.49 and 3.55 (2H, ABq, J18.4Hz), 3.96-4.00 (1H, m), 4.51-4.55 (1H, m), 4.72 (1H, d, J5.0Hz) and 4.93 (1H, d, J5.0Hz). [Mass spectrum:  $\underline{M}^+$  (340)]. The second isomer to be eluted (isomer B) was obtained as a white foam (400mg, 49%); 30  $\nu_{\text{max}}$  ( $\text{CH}_2\text{Cl}_2$ ), 1715, 1721 $\text{cm}^{-1}$ ;  $\delta_{\text{H}}$  ( $\text{CDCl}_3$ ) 1.56 (9H, s) overlapping 1.49-1.66 (4H, m), 1.84-2.05 (2H, m), 3.44 and 3.62 (2H, ABq, J 17.8) overlapping 3.45-3.54 (1H, m), 4.01-4.11 (1H, m), 4.56-4.61 (1H, m), 4.69 (1H, d, J5.0Hz) and 4.93 (1H, d, J5.0Hz). [Mass spectrum:  $\underline{M}^+$  (340)].

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(h) t-Butyl (6R,7R)-7-[2-(Z)-Methoxyimino-2-(2-tritylaminothiazol-4-yl)acetamido]-3-[tetrahydropyran-2-yl]ceph-3-em-4-carboxylate

5 Mesyl chloride (121mg, 82 $\mu$ l) was added to 2-(Z)-methoxyimino-2-(2-tritylaminothiazol-4-yl)acetic acid hydrochloride (466mg) and N,N-diisopropylethylamine (252mg, 340 $\mu$ l) in dry DMF (10ml) at -50°C under argon and stirred at -50°C for 1h. Then t-butyl (6R,7R)-7amino-3-(tetrahydropyran-2-yl)-10 ceph-3-em-4-carboxylate (Isomer A, 300mg) in dry DMF (5ml) followed by pyridine (70mg, 72 $\mu$ l) were added and reaction mixture left for a further 1h whilst warming to ambient temperature. The reaction mixture was partitioned between ethyl acetate and water, reextracted with ethyl acetate, 15 organic extracts washed with water (x3) and brine, dried, concentrated and flash chromatography (eluting with 30,40,50,60% ethyl acetate in hexane) to afford the title compound as a pale yellow foam (420mg, 62%);  $\nu_{\text{max}}$  (CH<sub>2</sub>Cl<sub>2</sub>) 3420, 1784, 1732 (shoulder), 1717, 1685cm<sup>-1</sup>;  $\delta_{\text{H}}$ (CDCl<sub>3</sub>), 1.53 20 (9H, s) overlapping 1.4-1.7 (4H, m), 1.73-1.94 (2H, m), 3.38-3.58 (3H, m), 3.95-4.00 (1H, m), 4.07 (3H, s), 4.54-4.59 (1H, m), 5.02 (1H, d, J4.8Hz), 5.90 (1H, dd, J4.5, 9.1Hz) 6.74 (1H, s), 6.86 (1H, d, J8.8Hz), 7.04 (1H, s) and 7.30 (15H, s). [Mass spectrum: +ve ion (3-nitrobenzyl 25 alcohol, sodium acetate) MNa<sup>+</sup> (788)].

(i) Sodium (6R,7R)-7-[2-(2-Aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(RS)-tetrahydropyran-2-yl]ceph-3-em-4-carboxylate

30 t-Butyl (6R,7R)-7-[2-(Z)-methoxyimino-2-(2-tritylaminothiazol-4-yl)acetamido]-3-(tetrahydropyran-2-yl)ceph-3-em-4-carboxylate (400mg) was dissolved in 0.1M hydrochloric acid in 90% formic acid (5.22ml) and set aside for 0.5h, 35 concentrated hydrochloric acid (50 $\mu$ l) added and left for a

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further 1.5h. The mixture was evaporated in vacuo, diluted with water, adjusted to pH6.7 with sodium bicarbonate, then chromatographed on HP20SS eluting with water then 1,2,4,6,8% THF in water. Fractions containing a diastereoisomeric mixture of the title compound (h.p.l.c.) were concentrated in vacuo and freeze dried (170mg, 66%);  $\nu_{\text{max}}$  (KBr) 1770, 1670, 1600, 1535cm<sup>-1</sup>;  $\delta_{\text{H}}$ [(CD<sub>3</sub>)<sub>2</sub>SO], 1.3-1.5 (4H, m), 1.6-1.85 (2H, m), 3.24-3.44 (m, masked by HOD peak), 3.83 (3H, s) overlapping 3.76-3.95 (1H, m), 4.46-4.50, 4.82-4.86 (together 1H, 2m), 4.94 (1H, d, J4.7Hz), 5.46-5.53 (1H, m), 6.74, 6.75 (together 1H, 2s), 7.23 (2H, s) and 9.48, 9.51 (together 1H, 2d, J5.6, 5.5Hz). [Mass spectrum: +ve ion (thioglycerol)  $\text{MH}^+$  (490),  $\text{MNa}^+$  (512)].

15 The second isomer eluted (isomer B) in step (g) (400mg) was progressed through step (h) as before yielding a pale yellow foam (550mg, 61%);  $\nu_{\text{max}}$ (CH<sub>2</sub>Cl<sub>2</sub>), 3420, 1783, 1729, 1687cm<sup>-1</sup>;  $\delta_{\text{H}}$ (CDCl<sub>3</sub>) 1.55 (9H, s) overlapping 1.44-1.68 (4H, m), 1.82-1.96 (2H, m), 3.44 and 3.65 (2H, ABq, J18.0) 20 overlapping 3.42-3.58 (1H, m), 4.07 (3H, s) overlapping 3.96-4.10 (1H, m), 4.66-4.69 (1H, m), 4.66-4.69 (1H, m), 5.01 (1H, d, J4.7Hz), 5.86 (1H, dd, J4.8, 8.9Hz), 6.75 (1H, s) overlapping 6.75-6.78 (1H, m), 7.01 (1H, s) and 7.30 (15H, s). [Mass spectrum: +ve ion (3-nitrobenzyl alcohol, 25 sodium acetate)  $\text{MNa}^+$  (788)]. This was then progressed through step (i) to afford the same mixture of diastereoisomers.

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Example 4

Pivaloyloxymethyl (6R,7R)-7-[2-(2-Aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(RS)-tetrahydropyran-2-yl]ceph-3-  
5 em-4-carboxylate

The title compound was prepared from the compound of Example 3 as described in Example 2 and obtained as a pale yellow foam (59%);  $\nu_{\text{max}}$  (CH<sub>2</sub>Cl<sub>2</sub>): 3388, 1787, 1752, 1688 cm<sup>-1</sup>;  
10  $\delta_{\text{H}}$  (CDCl<sub>3</sub>) 1.24 (9H, s), 1.42-1.64 (4H, m), 1.74-1.90 (2H, m), 3.40-3.75 (3H, m), 4.07, 4.08 (together 3H, 2s) overlapping 3.96-4.10 (1H, m), 4.56-4.59, 4.80-4.83 (together 1H, 2m), 5.07, 5.08 (together 1H, 2d, J4.8, 4.7Hz), 5.66 (2H, br.s), 5.85-6.03 (3H, m), 6.86, 6.89 15 (together 1H, 2s) and 7.59 (1H, d, J8.9Hz). [Mass spectrum: +ve ion (thioglycerol)  $\text{MH}^+$  (582);  $\text{MNa}^+$  (604)].

Example 5

20 (6R,7R)-7-[2-(2-Aminothiazol-4-yl)-2-(Z)-hydroxyiminoacet-  
amido]-3-[(RS)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylic  
acid

(a) t-Butyl (6R,7R)-3-[(R)-tetrahydrofuran-2-yl]-7-[2-  
25 (2-tritylaminothiazol-4-yl)-2-(Z)-trityloxyiminoacetamido]-  
ceph-3-em-4-carboxylate

Methanesulphonyl chloride (96 $\mu$ l, 1.25mmol) was added to sodium 2-(2-tritylaminothiazol-4-yl)-2-(Z)-trityloxy-  
30 iminoacetate (852mg, 1.2mmol) in DMF (2ml) at <-40°C. The mixture was stirred 0.5h at -30±10°C then t-butyl (6R,7R)-7-amino-3-[(R)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate (326mg, 1mmol) in DMF (2ml), followed by pyridine (101 $\mu$ l, 1.25mmol), were added. The reaction was stirred for 1h

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without further cooling then diluted with ethyl acetate, washed twice with water and with brine, dried, concentrated in vacuo and flash chromatographed eluting with 25, 30% ethyl acetate in hexane to give the title compound as a 5 colourless foam (665mg, 68%);  $\nu_{\text{max}}$  ( $\text{CH}_2\text{Cl}_2$ ) 3395, 1787, 1722, 1687, 1527, 1449, 1156 and  $1051\text{cm}^{-1}$ ;  $\delta_{\text{H}}$  ( $\text{CDCl}_3/\text{CD}_3\text{OH}$ ) 1.55 (9H, s), 1.65-2.25 (4H, m), 3.32 and 3.40 (2H, ABq, J 17.6Hz), 3.8-4.0 (2H, m), 5.08 (1H, d, J 4.8Hz), 5.13 (1H, dd, J 7.0, 8.1Hz), 5.91 (1H, d, J 4.5Hz), 6.56 (1H, s), 7.2-10 7.5 (30H, m). [Mass spectrum: +ve ion (3-nitrobenzyl alcohol, sodium acetate)  $\text{MNa}^+$  (1002)].

(b) (6R, 7R)-7-[2-(2-Aminothiazol-4-yl)-2-(Z)-hydroxy-iminoacetamido]-3-[(RS)-tetrahydrofuran-2-yl]ceph-3-em-4-15 carboxylic acid

t-Butyl (6R, 7R)-3-[(R)-tetrahydrofuran-2-yl]-7-[2-(2-tritylaminothiazol-4-yl)-2-(Z)-trityloxyiminoacetamido]ceph-3-em-4-carboxylate (660mg) was dissolved in 0.1M 20 hydrochloric acid in 90% formic acid (7ml) and left for 1h then concentrated hydrochloric acid (250 $\mu$ l) was added and left a further 0.75h. The mixture was evaporated to dryness in vacuo, the residue diluted with water, adjusted to pH3.2 with 0.25M sodium hydroxide then chromatographed on HP20SS 25 eluting with 0 to 15% THF in water. The fractions containing the title compound (h.p.l.c. analysis) were combined, concentrated and freeze-dried to give a colourless solid (102mg, 35%);  $\nu_{\text{max}}$  ( $\text{CH}_2\text{Cl}_2$ ) 3315, 1763, 1663, 1626, 1178 and  $1045\text{cm}^{-1}$ ;  $\delta_{\text{H}}$  [ $(\text{CD}_3)_2\text{SO}$ ] 1.6-2.2 (4H, m), 3.35-3.9 30 (4H, m), 4.73, 4.95 (together 1H, 2dd, J 8.3, 9.1Hz), 5.13, 5.15 (together 1H, 2d, J 4.6Hz), 5.7-5.8 (1H, m), 6.66, 6.68 (together 1H, 2s), 7.14 (2H, s), 9.44, 9.48 (1H, 2d, J 7.9Hz), 11.30, 11.31 (together 1H, 2s). [Mass spectrum: +ve ion (thioglycerol)  $\text{MH}^+$  (440)].

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Example 6

Diastereoisomers of (6R,7R)-7-Amino-3-(tetrahydrofuran-2-yl)ceph-3-em-4-carboxylate

5

(a) (RS)-2-Bromoacetyl tetrahydrofuran

A stream of diazomethane (from N-methyl-N-nitrosotoluene-4-sulphonamide, 18.0g) in argon (P. Lombardi, Chem. and Ind., 10 1990, (21), 708) was passed into a solution of (RS)-tetrahydrofuroyl chloride [prepared from (RS)-tetrahydrofuroic acid (3.48g, 30mmol) as described in Example 2(a)] in dichloromethane (60ml) cooled in an ice bath. When the diazomethane addition was complete, 48% 15 aqueous hydrogen bromide (5.6ml, 33.2mmol) was added. The mixture was stirred 0.25h then washed twice with water, dried, concentrated and flash chromatographed on silica gel eluting with 10% ethyl acetate in hexane to give the title compound as a pale yellow oil (4.44g, 77%);  $\nu_{\text{max}}$  ( $\text{CH}_2\text{Cl}_2$ ) 1733, 1245, 1073 and  $938\text{cm}^{-1}$ ;  $\delta_{\text{H}}$  ( $\text{CDCl}_3$ ) 1.85-2.35 (4H, m), 3.85-4.05 (2H, m), 4.20 (2H, s), 4.54 (1H, dd,  $J$  6.1, 8.2Hz). [Mass spectrum: +ve ion (ammonia)  $\text{MNH}_4^+$  (210)].

(b) 4-Methoxybenzyl (2RS)-2-hydroxy-2-[(3R,4R)-3-phenyl-acetamido-4-[(RS)-tetrahydrofuran-2-yl-carbonylmethylthio]azetidin-2-on-1-yl]acetate

Toluene-4-sulphonic acid (6.0g, 31.5mmol) in water (15ml) was added to a solution of 4-methoxybenzyl (2RS)-2-hydroxy-30 2-[(1R,5R)-3-benzyl-4-thia-2,6-diazabicyclo [3.2.0]hept-2-en-7-on-6-yl]acetate (7.42g, 18.0mmol) prepared from Penicillin G as described for the benzhydryl ester derived from Penicillin V, S. Yamamoto, N. Haga, T. Aoki, S. Hayashi, H. Tanida, and W. Nagata, Heterocycles, 1977, 8, 35 283) in dichloromethane (30ml) and acetone (30ml). After

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stirring for 2.5h at room temperature, the reaction mixture was diluted with dichloromethane, washed with water (x2), dried and concentrated in vacuo to yield crude 4-methoxybenzyl (2<sub>RS</sub>)-2-hydroxy-2-[(3<sub>R</sub>, 4<sub>R</sub>)-4-mercaptop-3-5 phenylacetamidoazetidin-2-on-1-yl]acetate as a yellow foam.

The crude thiol was dissolved in acetone (35ml) and treated with a solution of (RS)-2-bromoacetyltetrahydrofuran (3.48g, 18.0mmol) in acetone (5ml). After 10min, potassium 10 carbonate (1.24g, 8.9mmol) was added, and the mixture stirred for a further 30min. The reaction mixture was diluted with ethyl acetate, washed successively with water (x2) and brine, dried and concentrated. The residue was flash chromatographed on silica gel eluting with 50, 70 and 15 80% ethyl acetate in hexane yielding the title compound as a colourless foam (5.40g, 55%);  $\nu_{\text{max}}$  (CH<sub>2</sub>Cl<sub>2</sub>) 3409, 1781, 1745, 1684, 1613, 1516cm<sup>-1</sup>. [Mass spectrum +ve ion (3-nitrobenzyl alcohol, sodium acetate)  $\text{MNa}^+$  (565)].

20 (c) 4-Methoxybenzyl 2-[(3R, 4R)-3-phenylacetamido-4-[(RS)-tetrahydrofuran-2-ylcarbonylmethylthio]azetidin-2-on-1-yl]-2-tri-n-butylphosphoranylideneacetate

A solution of thionyl chloride (1.36ml, 18.6mmol) in THF 25 (10ml) was added dropwise to the hydroxy compound (6.72g, 12.4mmol) and 2,6-lutidine (2.16ml, 18.6mmol) in THF (30ml) at -20°C. After stirring for 1h, the reaction mixture was filtered through a pad of celite, and the filtrate evaporated in vacuo. Toluene was added and re-evaporated to 30 yield 4-methoxybenzyl (RS)-2-chloro-2-[(3<sub>R</sub>, 4<sub>R</sub>)-3-phenylacetamido-4-[(RS)-tetrahydrofuran-2-ylcarbonylmethylthio]azetidin-2-on-1-yl]acetate as an oil.

The crude chloro compound was dissolved in dioxan (30ml) and 35 treated with tri-n-butylphosphine (6.8ml, 27.3mmol). After

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stirring for 30min. at room temperature, the reaction mixture was diluted with ethyl acetate and washed successively with dilute sodium hydrogen carbonate solution, water and brine. The organic solution was dried, 5 concentrated and then flash chromatographed on silica gel eluting with 50, then 80% ethyl acetate in hexane to give the title compound as a foam (6.54g, 73%);  $\nu_{\text{max}}$  (CH<sub>2</sub>Cl<sub>2</sub>) 3422, 1763, 1732, 1680, 1613, 1515, 1174cm<sup>-1</sup>. [Mass spectrum +ve ion/thioglycerol)  $\text{M}^+$  (727),  $\text{MNa}^+$  (749)].

10

(d) 4-Methoxybenzyl (6R, 7R)-7-phenylacetamido-3-[(RS)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate

A solution of the phosphorane (6.40g, 8.82mmol) and benzoic acid (20mg) in toluene (100ml) was heated in an oil bath at 130°C for 10h under argon. The reaction mixture was cooled, 15 concentrated and the residue purified by chromatography on silica gel eluting with 20, 30, 40, 50% ethyl acetate in hexane yielding the title compound as a yellow oil (3.50g, 78% yield);  $\nu_{\text{max}}$  (CH<sub>2</sub>Cl<sub>2</sub>) 3411, 1783, 1723, 1688, 1515cm<sup>-1</sup>;  $\delta_{\text{H}}$  (CDCl<sub>3</sub>, 250MHz) 1.50-2.39 (together 4H, m), 3.27 and 3.60, 3.32 and 3.49 (together 2H, 2ABq, J 18.7, 17.9Hz), 20 3.58 and 3.70, 3.63 and 3.73 (together 2H, 2ABq, J 16.2, 16.1Hz), 3.80, 3.82 (together 3H, 2S), 3.84-3.97 (together 2H, 2m), 4.91, 5.18 (together 1H, 2m), 4.90, 4.94 (together 1H, 2d, J 4.7Hz), 5.17 (2H, s), 5.73, 5.82 (together 1H, 2dd, J 9.1, 4.7Hz), 5.98, 6.02 (together 1H, 2d, J 9.1Hz), 25 6.87 and 6.90 (together 2H, 2d, J 8.6Hz), 7.23-7.40 (7H, m). [Mass spectrum +ve ion (3-nitrobenzylalcohol, sodium acetate)  $\text{MNa}^+$  (531)].

(e) 4-Methoxybenzyl (6R, 7R)-7-amino-3-(tetrahydrofuran-2-yl)ceph-3-em-4-carboxylate

35 Phosphorus pentachloride (2.15g, 10.32mmol) in dichloromethane (108ml) was added to 4-methoxybenzyl

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(6R, 7R)-7-phenylacetamido-3-[(RS)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate (3.40g, 6.69mmol) and N-methylmorpholine (1.50ml, 13.64mmol) in dichloromethane (30ml) at -25°C. The reaction was stirred at -10 $\pm$ 5°C for 45min., then methanol 5 (14ml) was added, and stirring continued for 45min. at room temperature. Water (27ml) was then added, and the mixture vigorously stirred for a further 1h. The dichloromethane was evaporated in vacuo, the aqueous residue washed with diethyl ether, then adjusted to pH7 with ammonium hydroxide 10 in the presence of ethyl acetate. The mixture was extracted with ethyl acetate (x2), dried and concentrated in vacuo. The residue was purified by chromatography on silica gel eluting with 50, 70, 80% ethyl acetate in hexane yielding 4-methoxybenzyl (6R, 7R)-7-amino-3-[(S)-tetrahydrofuran-2- 15 yl]ceph-3-em-4-carboxylate (980mg, 38%) as a yellow foam;  $\nu_{\text{max}}$  (CH<sub>2</sub>Cl<sub>2</sub>) 1777, 1721, 1613, 1516, 1152cm<sup>-1</sup>;  $\delta_{\text{H}}$  (CDCl<sub>3</sub>, 250MHz) 1.53-1.71 (1H, m), 1.84-2.02 (4H, m, 2 exch.), 2.25-2.40 (1H, m), 3.31 and 3.60 (2H, ABq, J 18.5Hz), 3.78-3.98 (2H, m), 3.81 (3H, s), 4.72 (1H, d, J 5.0Hz), 4.86-4.93 (2H, 20 m), 5.19 (2H, s), 6.88 (2H, d, J 8.6Hz), 7.33 (2H, d, J 8.6Hz). [Mass spectrum: M<sup>+</sup> (390)].

Further elution with ethyl acetate gave the more polar diastereoisomer, 4-methoxybenzyl (6R, 7R)-7-amino-3-[(R)- 25 tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate (590mg, 23%). The product was recrystallised using ethyl acetate-hexane to yield an off-white solid m.p. 131-134°C.  $\nu_{\text{max}}$  (CH<sub>2</sub>Cl<sub>2</sub>) 1775, 1726, 1613, 1516, 1156cm<sup>-1</sup>;  $\delta_{\text{H}}$  (CDCl<sub>3</sub>, 250MHz) 1.58-1.70 (1H, m), 1.83-2.06 (4H, m, 2 exch.), 3.38 and 3.57 (2H, 30 ABq, J 17.8Hz), 3.77-3.93 (2H, m), 3.82 (3H, s), 4.68 (1H, d, J 4.9Hz), 4.92 (1H, d, J 4.9Hz), 5.07 (1H, m), 5.22 (2H, s), 6.90 (2H, d, J 8.6Hz), 7.35 (2H, d, J 8.6Hz). [Mass spectrum: M<sup>+</sup> (390)].

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Example 7

Sodium (6R, 7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(S)-tetrahydrofuran-2-yl]ceph-3-em-4-5 carboxylate

(a) 4-Methoxybenzyl (6R, 7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(S)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate

10 Methanesulphonyl chloride (203 $\mu$ l, 2.62mmol) was added to 2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetic acid (528mg, 2.63mmol) and N,N-diisopropylethylamine (458 $\mu$ l, 2.63mmol) in DMF (8ml) at -30 $^{\circ}$ C. After stirring at -30 $\pm$ 10 $^{\circ}$ C for 30min., 15 a solution of 4-methoxybenzyl (6R, 7R)-7-amino-3-[(S)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate (930mg, 2.38mmol) in DMF (5ml) was added, followed by pyridine (213 $\mu$ l, 2.63mmol). The reaction mixture was transferred to an ice-bath and stirring continued for a further 1h. After 20 dilution with ethyl acetate, the solution was washed successively with saturated sodium hydrogen carbonate solution, 5% aqueous citric acid, water (x2) and brine, dried and then concentrated in vacuo. The residue was purified by chromatography on silica gel eluting with 50, 70 25 and 90% ethyl acetate in hexane to give the title compound as a yellow foam (1.138g, 83%);  $\nu_{\text{max}}$  (CH<sub>2</sub>Cl<sub>2</sub>) 3389, 1783, 1732, 1682, 1516cm<sup>-1</sup>;  $\delta_{\text{H}}$  (CDCl<sub>3</sub>, 250MHz) 1.53-1.70 (1H, m), 1.88-2.01 (2H, m), 2.28-2.41 (1H, m), 3.33 and 3.62 (2H, ABq, J 18.7Hz), 3.79-3.98 (2H, m), 3.81 (3H, s), 4.08 (3H, s), 4.94 (1H, dd, J 9.0, 6.7Hz), 5.04 (1H, d, J 4.8Hz), 5.18 (2H, s), 5.88 (2H, br s, exch.), 5.98 (1H, dd, J 9.0, 4.8Hz), 6.90 (2H, d, J 8.6Hz), 6.94 (1H, s), 7.35 (2H, d, J 8.6Hz), 7.50 (1H, br. d, J 9.0Hz, exch.). [Mass spectrum: +ve ion (thioglycerol)  $\text{MH}^+$  (574)].

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(b) Sodium (6R,7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(S)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate

5 Aluminium chloride (162mg, 1.21mmol) was added to anisole (7ml) and dry dichloromethane (3.5ml) at -20°C and stirred for 15min. The temperature of the cooling bath was then lowered to -40°C before addition of a solution of 4-methoxybenzyl (6R,7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(S)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate (235mg, 0.41mmol) in dichloromethane (5ml). After 10min., the solution was treated with trisodium citrate (0.5M, 12ml) and then vigorously stirred for 10min at room temperature. The aqueous phase was separated, 15 washed with dichloromethane (x2) and concentrated in vacuo. The residue was chromatographed on HP20SS eluting with water, then 1% THF in water. Fractions containing the product, (h.p.l.c. analysis), were combined and freeze-dried to give the title compound (126mg, 65%);  $\nu_{\text{max}}$  (KBr) 3401, 1761, 1669, 1603, 1533, 1040cm<sup>-1</sup>;  $\delta_{\text{H}}$  (d<sub>6</sub>-DMSO, 250MHz) 1.43-1.59 (1H, m), 1.71-1.88 (2H, m), 2.0-2.12 (1H, m), 3.18 and 3.37 (2H, ABq, J 17.4Hz), 3.58 (1H, m), 3.78 (1H, m), 3.81 (3H, s), 4.87 (1H, dd, J 8.7, 6.7Hz), 4.97 (1H, d, J 4.7Hz), 5.50 (1H, dd, J 8.1, 4.7Hz), 6.74 (1H, s), 7.21 (2H, br. s, exch.), 9.48 (1H, d, J 8.1Hz, exch.). [Mass spectrum: +ve ion (thioglycerol)  $\text{MH}^+$  (476),  $\text{MNa}^+$  (498)].

Example 8

30

Pivaloyloxymethyl (6R,7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(S)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate

35 Pivaloyloxymethyl bromide (440mg, 2.26mmol) and sodium iodide (440mg, 2.93mmol) in acetone (3ml) were stirred for

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30min., filtered, and the filtrate concentrated in vacuo. The resulting iodide in toluene (2ml) was added to a solution of sodium (6R, 7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[ (S)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate (560mg, 1.18mmol) in N-methylpyrrolidinone (5ml). After stirring for 45min. at room temperature, the reaction mixture was diluted with ethyl acetate, washed successively with water (x3) and brine, dried over MgSO<sub>4</sub> and concentrated in vacuo. The residue was chromatographed on 10 silica gel eluting with 80% ethyl acetate in hexane to give the title compound as a yellow foam (486mg, 73%);  $\nu_{\text{max}}$  (CH<sub>2</sub>Cl<sub>2</sub>) 3390, 1776, 1749, 1681, 1532cm<sup>-1</sup>;  $\delta_{\text{H}}$  (CDCl<sub>3</sub>, 250MHz) 1.22 (9H, s), 1.65 (1H, m), 1.99 (2H, m), 2.41 (1H, m), 3.37 and 3.68 (2H, ABq, J 18.8Hz), 3.80-4.01 (2H, m), 15 4.13 (3H, s), 4.92 (1H, dd, J 8.9, 6.8Hz), 5.08 (1H, d, J 4.8Hz), 5.85 and 5.92 (2H, ABq, J 5.6Hz), 5.98 (1H, dd, J 8.4, 4.8Hz), 6.07 (2H, br s, exch.), 7.03 (1H, s), 7.40 (1H, br. d, exch. J 8.4Hz). [Mass spectrum: +ve ion (thioglycerol) MH<sup>+</sup> (568)].

20

Example 9

Sodium (6R, 7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[ (R)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate

(a) 4-Methoxybenzyl (6R, 7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[ (R)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate

30

Methanesulphonyl chloride (198 $\mu$ l, 2.56mmol) was added to 2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetic acid (515mg, 2.56mmol) and N,N-diisopropylethylamine (447 $\mu$ l, 2.57mmol) in DMF (8ml) at -30°C. After stirring at -30±10°C for 30min., 35 a solution of 4-methoxybenzyl (6R, 7R)-7-amino-3-[ (R)-

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tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate (915mg, 2.35mmol) in DMF (5ml) was added, followed by pyridine (207 $\mu$ l, 2.56mmol). The reaction mixture was transferred to an ice-bath and stirring continued for a further 1.5h:  
5 After dilution with ethyl acetate, the solution was washed successively with saturated sodium hydrogen carbonate solution, 5% aqueous citric acid, water (x2) and brine, dried and then concentrated in vacuo. The residue was triturated several times with diethyl ether to yield the  
10 title compound as an off-white solid (1.06g, 79%);  $\nu_{\text{max}}$  ( $\text{CH}_2\text{Cl}_2$ ) 3390, 1783, 1730, 1687, 1606, 1516 $\text{cm}^{-1}$ ;  $\delta_{\text{H}}$  ( $\text{CDCl}_3$ , 250MHz) 1.55-1.70 (1H, m), 1.86-1.98 (2H, m), 2.0-2.14 (1H, m), 3.40 and 3.59 (2H, ABq, J 17.8Hz), 3.78-3.93 (2H, m), 3.91 (3H, s), 4.12 (3H, s), 5.04 (1H, d, J 4.7Hz), 5.15 (1H, dd, J 7.7, 7.7Hz), 5.21 (2H, s), 5.87 (1H, dd, J 8.7, 4.7Hz), 6.55 (2H, br. s, exch.), 6.90 (2H, d, J 8.6Hz), 7.05 (1H, s), 7.36 (2H, d, J 8.6Hz), 7.65 (1H, br. d, J 8.7Hz).  
[Mass spectrum: +ve ion (thioglycerol)  $\text{M}^+$  (574)].

20 (b) Sodium (6R,7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(R)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate

Aluminium chloride (740mg, 5.55mmol) was added to anisole 25 (32ml) and dry dichloromethane (15ml) at -20°C and stirred for 15min. The temperature of the cooling bath was then lowered to -40°C before addition of a solution of 4-methoxybenzyl (6R,7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(R)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate (1.06g, 1.85mmol) in dichloromethane (10ml). After 10min., the solution was treated with trisodium citrate (0.5M, 54ml) and then vigorously stirred for 10min. at room temperature. The aqueous phase was separated, washed with dichloromethane (x2) and concentrated 35 in vacuo. The residue was chromatographed on HP20SS eluting with water, then 1% THF in water. Fractions containing the

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product, (h.p.l.c. analysis), were combined and freeze-dried to give the title compound (560mg, 64%);  $\nu_{\text{max}}$  (KBr) 3399, 1762, 1669, 1603, 1529, 1038cm<sup>-1</sup>;  $\delta_{\text{H}}$  ( $d_6$ -DMSO, 250MHz) 1.50-1.91 (4H, m), 3.25 and 3.38 (2H, ABq, J 16.8Hz), 3.60-3.82 (2H, m), 3.84 (3H, s), 4.96 (1H, d, J 4.7Hz), 5.20 (1H, dd, J 8.6, 6.0Hz), 5.48 (1H, dd, J 8.1, 4.7Hz), 6.76 (1H, s), 7.23 (2H, br. s, exch.), 9.50 (1H, d, J 8.1Hz, exch.). [Mass spectrum: +ve ion (thioglycerol)  $\text{MH}^+$  (476),  $\text{MNa}^+$  (498)].

10

Example 10

Pivaloyloxymethyl (6R,7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(R)-tetrahydrofuran-2-yl]ceph-3-  
15 em-4-carboxylate

Pivaloyloxymethyl bromide (247mg, 1.27mmol) and sodium iodide (247mg, 1.65mmol) in acetone (5ml) were stirred for 30min., filtered, and the filtrate concentrated in vacuo.

20 The resulting iodide in toluene (3ml) was added to a solution of sodium (6R, 7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(R)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate (320mg, 0.67mmol) in N-methylpyrrolidinone (5ml). After stirring for 30min. at room temperature, the 25 reaction mixture was diluted with ethyl acetate, washed successively with water (x3) and brine, dried over  $MgSO_4$  and concentrated in vacuo. The residue was chromatographed on silica gel eluting with 80% ethyl acetate in hexane to give the title compound as a yellow foam (297mg, 78%);  $\nu_{max}$

30  $(CH_2Cl_2)$  3387, 1786, 1752, 1735, 1686, 1605 $cm^{-1}$ ;  $\delta_H$  ( $CDCl_3$ , 250MHz) 1.22 (9H, s), 1.69 (1H, m), 1.98 (2H, m), 2.18 (1H, m), 3.43 and 3.62 (2H, ABq,  $J$  18.0Hz), 3.80-3.96 (2H, m), 4.10 (3H, s), 5.08 (1H, d,  $J$  4.7Hz), 5.19 (1H, m), 5.83-5.92 (3H, m), 6.32 (2H, br. s, exch), 7.02 (1H, s), 7.63 (1H, br. d, exch.,  $J$  8.6Hz). [Mass spectrum: +ve ion (thioglycerol)  $MH^+$  (568)].

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Example 11

Diphenylmethyl (6R,7R)-7-phenylacetamido-3-[(RS)-tetra-  
hydrofuran-2-yl]ceph-3-em-4-carboxylate

5

(a) Diphenylmethyl (6R,7R)-7-phenylacetamido-3-(tetra-  
hydrofuran-2-yl)ceph-2-em-4-carboxylate

A solution of (tetrahydrofuran-2-yl)tri-*n*-butylstannane  
10 (J.S. Sawyer, A. Kucerovy, T.L. MacDonald, and  
G.J. McGarvey, J. Amer. Chem. Soc., 1988, 110, 842) (3.0g,  
8.30mmol) in THF (20ml) was cooled to -78°C. *n*-Butyl  
lithium (6.23ml of a 1.6M solution in hexane, 9.97mmol) was  
then added and the solution was stirred for 15min. at -78°C.  
15 A second flask containing copper (I) bromide.dimethyl-  
sulphide complex (0.854g, 4.14mmol) suspended in a mixture  
of dimethyl sulphide (15ml) and THF (30ml) was then cooled  
to -78°C. The  $\alpha$ -lithiotetrahydrofuran species was  
transferred via a cannula to the suspension of copper  
20 bromide at -78°C. The red-brown homogeneous solution was  
stirred for 30min. at -78°C. A third flask containing a  
solution of diphenylmethyl 7-phenylacetamido-3-  
triflyloxyceph-3-em-4-carboxylate (V. Farina, S.R. Baker,  
and S.I. Hanck, J. Org. Chem., 1989, 54, 4962) (1.9g,  
25 3.0mmol) in a mixture of N-methylpyrrolidinone (20ml) and  
THF (50ml) was then cooled to -78°C. The cuprate species  
was transferred via a cannula to the solution of triflate at  
-78°C. The reaction mixture was stirred for 1h at -78°C  
then quenched by the addition of saturated aq. ammonium  
30 chloride (30ml). The resulting mixture was allowed to warm  
to room temperature then diluted with water (100ml) and  
extracted with ethyl acetate (100ml, 30ml). The combined  
organic phases were washed with water, brine, then dried  
over magnesium sulphate. After removal of the solvents  
35 under reduced pressure the crude reaction product was  
purified by flash chromatography on silica gel using 10-30%  
ethyl acetate/methylene dichloride as eluent. After elution

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of the 3-n-butylcephem, the title compound was obtained as a mixture of diastereoisomers of the  $\Delta^2$  and  $\Delta^3$  cephems (1.014g, 61%).

5 (b) Diphenylmethyl (6R,7R)-1-oxo-7-phenylacetamido-3-(tetrahydrofuran-2-yl)ceph-3-em-4-carboxylate

A mixture of the cephems (1.014g, 1.83mmol) obtained in Example 11(a) in methylene dichloride (20ml) was cooled to 10  $0^{\circ}\text{C}$ . A solution of m-chloroperbenzoic acid (0.52g, 60% pure, 1.81mmol) in methylene dichloride (10ml) was then added and the solution was stirred for 10min. at  $0^{\circ}\text{C}$ . The solution was washed with saturated aq. sodium hydrogen carbonate then water and dried ( $\text{MgSO}_4$ ). Evaporation of the 15 solvent gave the title compound (1.005g, 96%) as a mixture of diastereoisomers;  $\nu_{\text{max}}$  (KBr) 1786, 1728 and  $1648\text{cm}^{-1}$ ;  $\delta$  ( $\text{CDCl}_3$ ) 1.41-2.29 (4H, m), 2.99, 3.27 (together 1H, 2d,  $J$  19Hz), 3.63 (2H, br. s), 3.63-3.87 (2.5H, m), 4.20 (0.5H, d), 4.41, 4.43 (together 1H, m), 4.97, 5.14 (together 1H, 20 br. t,  $J$  7.5Hz, and dd,  $J$ , 9, 6.9Hz), 6.05, 6.09 (together 1H, 2dd,  $J$  10, 4.7Hz), 6.70, 6.82 (together 1H, 2d,  $J$  10.1Hz), 6.87, 6.94 (together 1H, 2s) and 7.26-7.40 (15H, m). [Mass spectrum: +ve ion (3-nitrobenzyl alcohol, sodium acetate)  $\text{MNa}^+$  (593)].

25

(c) Diphenylmethyl (6R,7R)-7-phenylacetamido-3-(tetrahydrofuran-2-yl)ceph-3-em-4-carboxylate

A solution of the sulphoxides (0.975g, 1.71mmol) obtained in 30 Example 11(b) in DMF (20ml) was cooled to  $-25^{\circ}\text{C}$ . Phosphorous trichloride (0.30ml, 3.44mmol) was then added and the solution was stirred for 10min. at  $-25^{\circ}\text{C}$ . The reaction mixture was poured onto a mixture of ice, water and ethyl acetate. The organic extract was washed with water, 35 brine, dried ( $\text{MgSO}_4$ ) and evaporated. Purification by flash

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chromatography gave the title compound as a mixture of diastereoisomers (0.811g, 86%);  $\nu_{\text{max}}$  (KBr) 1780, 1723 and 1663  $\text{cm}^{-1}$ ;  $\delta$  ( $\text{CDCl}_3$ ) 1.5-2.3 (4H, m), 3.24 (0.5H, d,  $J$  18.6 Hz), 3.40 (0.5H, d,  $J$  17.3 Hz), 3.56-3.89 (5H, m), -4.84 s (0.5H, dd,  $J$  9.1, 6.7 Hz), 4.95 (1H, d,  $J$  4.8 Hz), 5.01 (0.5H, br t,  $J$  8 Hz) 5.76, 5.85 (together 1H, 2dd,  $J$  8.9, 4.8 Hz), 6.01, 6.08 (together 1H, 2d,  $J$  8.9 Hz), 6.86, 6.94 (1H, 2s) and 7.26-7.38 (15H, m) [mass spectrum:  $\underline{M}^+$  (554)].

10 Example 12

Pivaloyloxymethyl (6R,7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-(tetrahydrofuran-2-yl)ceph-3-em-4-carboxylate

15

(a) Pivaloyloxymethyl (6R,7R)-7-phenylacetamido-3-(tetrahydrofuran-2-yl)ceph-3-em-4-carboxylate

A solution of the cephems (0.811g, 1.46mmol) obtained in  
20 Example 11(c) in anisole (5ml) was cooled to 0°C.

Trifluoroacetic acid (10ml) was added and the mixture was stirred at 0°C for 5min. Toluene was added and the solvents were evaporated off. The residue was partitioned between water and ethyl acetate and the pH was adjusted to 7 by the 25 addition of saturated aq. sodium hydrogen carbonate. The aqueous layer was added to ethyl acetate and the pH taken to 2 by the addition of 1M HCl. The organic phase was washed with water, brine, dried ( $\text{MgSO}_4$ ) and evaporated. The residue was dissolved in N-methylpyrrolidinone (3ml).

30 Potassium carbonate (0.426g, 3.08mmol) was added followed by a solution of iodomethyl pivalate (prepared from the bromide 0.438g as in Example 2) in toluene (3ml). The mixture was stirred for 2h at room temperature, then water and ethyl acetate were added. The organic phase was washed with 35 water, brine, dried ( $\text{MgSO}_4$ ) and evaporated. The residue was

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purified by chromatography to give the title compound as a (5:1) mixture of diastereoisomers (0.478g, 65%); major diastereoisomer (S)  $\delta_{\text{H}}$  ( $\text{CDCl}_3$ ) 1.22 (9H, s), 1.56 (1H, m), 1.96 (2H, m), 2.35 (1H, m), 3.27 (1H, d,  $J$  18.8Hz), 3.60 (1H, d), 3.65 (2H, ABq,  $J$  16.2Hz), 3.88 (2H, m), 4.86 (1H, dd,  $J$  9.0, 6.7Hz), 4.94 (1H, d,  $J$  4.8Hz), 5.79-6.05 (4H, m) and 7.26-7.38 (5H, m).

(b) Pivaloyloxymethyl (6R,7R)-7-amino-3-(tetrahydro-10 furan-2-yl)ceph-3-em-4-carboxylate

A solution of the diastereoisomers obtained in Example 12(a) (0.478g, 0.95mmol) in methylene dichloride (10ml) was cooled to  $-30^{\circ}\text{C}$ . N-Methylmorpholine (0.206ml, 1.87mmol) was added 15 followed by a solution of phosphorus pentachloride (0.30g, 1.44mmol) in methylene dichloride (7.5ml). The mixture was stirred at  $-30^{\circ}\text{C}$  for 30min. Methanol (2.0ml) was added and the mixture was allowed to warm to room temperature over 30min. Water (2.6ml) was then added and the mixture was 20 stirred vigorously for 1h. The mixture was concentrated by evaporation under reduced pressure and the residue was partitioned between ethyl acetate and water. The pH was adjusted to 7 with 1M aq. ammonia. The organic phase was washed with water, brine, dried ( $\text{MgSO}_4$ ) and concentrated.

25 The diastereoisomers were separated by flash chromatography to give (S)-isomer (0.195g); (Found:  $\underline{M}^+$ , 384.1363.  $\text{C}_{17}\text{H}_{24}\text{N}_2\text{O}_6\text{S}$  requires  $\underline{M}$ , 384.1355);  $\nu_{\text{max}}$  ( $\text{KBr}$ ) 3408, 2977, 1780 and  $1750\text{cm}^{-1}$ ;  $\delta$  ( $\text{CDCl}_3$ ), 1.23 (9H, s), 1.64 (1H, m), 1.98 (2H, m), 2.10 (2H, br. s), 2.39 (1H, m), 3.35 (1H, d,  $J$  18.7Hz), 3.63 (1H, d,  $J$  18.6Hz), 3.90 (2H, m), 4.79 (1H, d,  $J$  5.0Hz), 4.88 (1H, dd,  $J$  9.1, 6.7Hz), 4.94 (1H, d,  $J$  5.0Hz) and 5.86 (2H, m). (R)-isomer (0.046mg);  $\delta$  ( $\text{CDCl}_3$ ) 1.23 (9H, s), 1.6-2.4 (6H, m), 3.43 (1H, d,  $J$  18Hz), 3.64 (1H, d,  $J$  17.6Hz), 3.88 (2H, m), 4.79 (1H, d,  $J$  4.9Hz), 4.99 (1H, d,  $J$  4.9Hz), 5.17 (1H, t,  $J$  7.5Hz) and 5.87 (2H, m).

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(c) Pivaloyloxymethyl (6R,7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(S)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate

5 A solution of (Z)-2-(2-aminothiazol-4-yl)-2-methoxyiminoacetic acid (0.108g, 0.537mmol) in DMF (2ml) was cooled to -50°C. N,N-Diisopropylethylamine (0.103ml, 0.59mmol) followed by methanesulphonyl chloride (0.046ml, 0.59mmol) were added and the mixture was stirred at -50°C for 30min.

10 A further quantity of N,N-diisopropylethylamine (0.086ml, 0.493mmol) was added and this mixture was added to a pre-cooled solution of pivaloyloxymethyl (6R,7R)-7-amino-3-[(S)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate (0.185g, 0.482mmol) in DMF (2ml) at 0°C. The resulting mixture was 15 stirred at 0°C for 40min., then it was partitioned between ethyl acetate and water. The organic phase was washed with water, brine, dried ( $MgSO_4$ ) and evaporated. The residue was purified by flash chromatography, then triturated with ether to give the title compound (0.193g, 71%) as a white solid.

20 The spectral data was identical with that obtained for Example 8.

(d) Pivaloyloxymethyl (6R,7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(R)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate

A solution of (Z)-2-(2-aminothiazol-4-yl)-2-methoxyiminoacetic acid (27mg, 0.134mmol) in DMF (1ml) was cooled to -50°C. N,N-Diisopropylethylamine (26 $\mu$ l, 0.15mmol) 30 followed by methanesulphonyl chloride (11.5 $\mu$ l, 0.15mmol) were added and the mixture was stirred at -50°C for 30min. A further quantity of N,N-diisopropylethylamine (22 $\mu$ l, 0.126mmol) was added and this mixture was added to a pre-cooled solution of pivaloyloxymethyl (6R,7R)-7-amino-3-[(R)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate (46mg, 0.12mmol) in DMF (1ml) at 0°C. The resulting mixture was

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stirred at 0°C for 40min., then it was partitioned between ethyl acetate and water. The organic phase was washed with water, brine, dried ( $MgSO_4$ ) and evaporated. The residue was purified by flash chromatography, then triturated with ether to give the title compound (49.6mg, 73%) as a white solid.

The spectral data was identical with that in Example 10.

Example 13

10 Sodium (6R, 7R)-7-[2-(2-Aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido-3-[ (RS)-tetrahydrofuran-3-yl]ceph-3-em-4-carboxylate

15 (a) (RS)-3-Chloroacetyltetrahydrofuran  
(RS)-3-Tetrahydrofuroic acid (3.48g) in dichloromethane (40ml) was treated with oxalyl chloride (11.43g) as described in Example 1(a). The resultant acid chloride in dichloromethane (40ml) was then treated with excess 20 diazomethane (60mM) in ether (100ml), followed by hydrogen chloride. The solution was washed once with brine, dried and concentrated. Flash chromatography on silica gel, eluting with 40% ethyl acetate/hexane afforded the title compound as a pale yellow oil, (3.924g, 88%);  $\nu_{max}$  ( $CH_2Cl_2$ ) 25 1735 and  $1716\text{cm}^{-1}$ ; 2.17 (2H, dt,  $J$  7.0, 7.5Hz), 3.47-3.58 (1H, m), 3.77-4.04 (4H, m) and 4.18 (2H, s); [mass spectrum: +ve ion (ammonia)  $MNH_4^+$  (166)].

30 (b) (3R, 4R)-3-Phenylacetamido-4-[tetrahydrofuran-3-ylcarbonylmethylthio]azetidin-2-one

(RS)-3-Chloroacetyltetrahydrofuran (0.297g) was coupled with (3R, 6R)-4-mercaptop-3-phenylacetamidoazetidin-2-one (0.519g) in DMF (4ml), using potassium carbonate (0.304g) as

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described in Example 1(b). Following work up, the crude product was taken up in hot ethyl acetate and cooled. The crystalline product was filtered off. The solvent was removed from the filtrate and the residue triturated with 5 dichloromethane. The crystalline products were combined to give one diastereoisomer of the title compound, (0.187g, 27%); m.p. 145-155°C (decomp.);  $\nu_{\text{max}}$  (CH<sub>2</sub>Cl<sub>2</sub>) 3410, 1748, 1709 (shoulder) and 1688cm<sup>-1</sup>;  $\delta_{\text{H}}$  ((CD<sub>3</sub>)<sub>2</sub>SO) 1.74-2.07 (2H, m), 3.26-3.38 (1H, m), 3.48 and 3.56 (2H, ABq, J 16.5Hz), 10 3.60-3.75 (4H, m), 4.87 (1H, d, J 4.5Hz), 5.24 (1H, dd, J 4.5, 8.4Hz collapses to 1H, d, J 4.5Hz with D<sub>2</sub>O) and 9.02 (1H, d, J 8.4Hz, exchangeable with D<sub>2</sub>O); [mass spectrum: +ve ion (3NOBA, Na<sup>+</sup>) MNa<sup>+</sup> (371)]. The dichloromethane soluble residue was flash chromatographed with ethyl acetate to give 15 the second diastereoisomer of the title compound as a colourless foam (0.162g, 23%);  $\nu_{\text{max}}$  (CH<sub>2</sub>Cl<sub>2</sub>) 3407, 3302 (br), 1783 and 1681cm<sup>-1</sup>;  $\delta_{\text{H}}$  ((CD<sub>3</sub>)<sub>2</sub>SO) spectrum identical to that of previous isomer except for 3.44-3.58 (2H, m); [mass spectrum: +ve ion (3NOBA, Na<sup>+</sup>) MH<sup>+</sup> (349), MNa<sup>+</sup> (371)].

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(c) t-Butyl (RS)-2-Hydroxy-2-[(3R,4R)-3-phenylacetamido-4-[(RS)-tetrahydrofuran-3-ylcarbonylmethylthio]azetidin-2-on-1-yl]acetate

25 t-Butyl glyoxylate (1.601g) in 1,2-dichloroethane (20ml) was added to (3R,4R-3-phenylacetamido-4-[(RS)-tetrahydrofuran-3-ylcarbonylmethylthio]azetidin-2-one (2.712g) with triethylamine (0.079g, 0.108ml) in 1,2-dichloroethane (5ml) at room temperature; after 1h. the solution was concentrated 30 and flash chromatographed with 70, 80 then 90% ethyl acetate/hexane to give the title compound as a colourless foam, (2.719g, 73%);  $\nu_{\text{max}}$  (CH<sub>2</sub>Cl<sub>2</sub>) 3415 (br), 1780, 1735, 1685 and 1509cm<sup>-1</sup>;  $\delta_{\text{H}}$  (CDCl<sub>3</sub>) 1.48 and 1.51 (9H, 2s's), 2.03-2.18 (2H, m), 3.20-3.32 (1H, m), 3.46 (1H, d, J 17.5Hz), 3.66 (2H, s), 3.69-3.97 (5H, m), 4.37 and 4.49 (1H, 2 br. d's, J 6.8 and 7.3Hz, exchangeable with D<sub>2</sub>O), 4.98 and

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5.05 (1H, 2d's, J 4.7 and 4.6Hz), 5.15-5.50 (2H, 4m's),  
6.43-6.74 (1H, 3m's) and 7.32 (5H, m); [mass spectrum: +ve  
ion (3NOBA, Na<sup>+</sup>), MNa<sup>+</sup> (501)].

5 (d) t-Butyl 2-[(3R,4R)-3-Phenylacetamido-4-[(RS)-  
tetrahydrofuran-3-ylcarbonylmethylthio]azetidin-2-on-1-yl]-  
2-tri-n-butylphosphoranylideneacetate

t-Butyl (RS)-2-hydroxy-2-[(3R,4R)-3-phenylacetamido-4-[(RS)-  
10 tetrahydrofuran-3-ylcarbonylmethylthio]azetidin-2-on-1-  
yl]acetate (2.719g) in THF (20ml) was treated with thionyl  
chloride (1.01g, 0.615ml) and 2,6-lutidine (0.913g, 0.989ml)  
as described in Example 1(d). Following work-up the crude  
chloride in dioxan (30ml) was then treated with n-butyl-  
15 phosphine (2.53g, 3.11ml). After purification by flash  
chromatography with 50, 70% ethyl acetate/hexane then ethyl  
acetate the title compound was obtained as a pale yellow  
foam (1.496g, 40%);  $\nu_{\text{max}}$  (CH<sub>2</sub>Cl<sub>2</sub>) 3420, 1762, 1717  
(shoulder), 1681 and 1625cm<sup>-1</sup>. [Mass spectrum: +ve ion  
20 (3NOBA, Na<sup>+</sup>), MH<sup>+</sup> (663), MNa<sup>+</sup> (685)].

(e) t-Butyl (6R,7R)-7-Phenylacetamido-3-[(RS)-tetra-  
hydrofuran-3-yl]ceph-3-em-4-carboxylate

25 t-Butyl 2-[(3R,4R)-3-phenylacetamido-4-[(RS)-  
tetrahydrofuran-3-ylcarbonylmethylthio]azetidin-2-on-1-yl]-  
2-tri-n-butylphosphoranylideneacetate (1.496g), thermolysed  
in toluene (30ml) as for Example 1(e) and purified by flash  
chromatography with 40, 50 and 60% ethyl acetate/hexane  
30 afforded the title compound as a yellow foam (0.28g, 28%);  
 $\nu_{\text{max}}$  (CH<sub>2</sub>Cl<sub>2</sub>) 3411, 1702, 1718 and 1687cm<sup>-1</sup>;  $\delta_{\text{H}}$  (CDCl<sub>3</sub>) 1.52  
(9H, s), 1.43-2.39 (3H, m's), 3.23 and 3.44 with 3.27 and  
3.44 (2H, 2ABq's J 17.7Hz), 3.51-4.03 (6H, m's), 4.94 and  
4.96 (1H, 2d's, J 4.7 and 4.7Hz), 5.74-5.82 (1H, m), 6.03

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and 6.04 (1H, 2d's,  $J$  8.8 and 8.9 Hz) and 7.26-7.42 (5H, m). [Mass spectrum: +ve ion (3NOBA,  $\text{Na}^+$ )  $\text{MNa}^+$  (467)].

(f) t-Butyl (6R, 7R)-7-Amino-3-[(RS)-tetrahydrofuran-3-yl]ceph-3-em-4-carboxylate

t-Butyl (6*R*, 7*R*)-7-phenylacetamido-3-[(*RS*)-tetrahydrofuran-3-yl]ceph-3-em-4-carboxylate (0.9g) in dichloromethane (15ml) with N-methylmorpholine (0.45g, 0.49ml) was successively 10 treated with phosphorus pentachloride (0.549g) in dichloromethane (13.74ml), methanol (10ml) and water (10ml) as described in Example 2(f). After purification by flash chromatography on silica gel eluting with 60, 80% ethyl acetate/hexane and then ethyl acetate, the title compound 15 was obtained as a yellow solid (0.481g, 73%); (Found:  $M^+$ , 326.1304.  $C_{15}H_{22}N_2O_4S$  requires  $M$ , 326.1300);  $\nu_{max}$  ( $CH_2Cl_2$ ) 3408, 1775 and  $1716\text{cm}^{-1}$ ;  $\delta_H$  ( $CDCl_3$ ) 1.55 (9H, s), 1.69-2.41 (3H, m's), 3.31 and 3.48 with 3.34 and 3.49 (2H, 2ABq's,  $J$  17.5 and 17.5Hz), 3.69-3.83 (4H, 2s's overlapping m), 3.97-20 4.05 (2H, m), 4.72 and 4.74 (1H, 2d's,  $J$  4.3 and 4.4Hz) and 4.95 and 4.97 (1H, 2d's,  $J$  4.3 and 4.4Hz).

(g) t-Butyl (6R, 7R)-7-[2-(Z)-Methoxyimino-2-(2-trityl-aminothiazol-4-yl)acetamido]-3-[(RS)-tetrahydrofuran-3-yl]ceph-3-em-4-carboxylate

2-(Z)-Methoxyimino-2-(2-tritylaminothiazol-4-yl)acetic acid hydrochloride (0.751g) in DMF (5ml) was treated with methanesulphonyl chloride (0.179g, 0.121ml) and 30 diisopropylethylamine (0.404g, 0.544ml) as described in Example 1(g). This was then treated with t-butyl (6R,7R)-7-amino-3-[(RS)-tetrahydrofuran-3-yl]ceph-3-em-4-carboxylate (0.464g) and pyridine (0.112g, 0.114ml) in DMF (5ml). Following work up and purification by flash chromatography 35 with 40, 50 and 60% ethyl acetate/hexane, the title compound

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was obtained as a yellow foam (0.874g, 82%);  $\nu_{\text{max}}$  (CH<sub>2</sub>Cl<sub>2</sub>) 3398, 1783, 1731 (shoulder), 1718 and 1688cm<sup>-1</sup>;  $\delta_{\text{H}}$  (CDCl<sub>3</sub>) 1.53 (9H, s), 1.69-2.43 (3H, m's), 3.29 and 3.46 with 3.34 and 3.48 (2H, 2ABq's, J 17.7 and 17.7Hz), 3.63-4.07 (6H, m's) 5 and s), 5.03 and 5.06 (1H, 2d's, J 4.8 and 4.8Hz), 5.84-5.90 (1H, m), 6.73 and 6.74 (1H, 2s), 6.76 and 6.90 (1H, 2d's, J 8.7 and 8.7Hz exchangeable with D<sub>2</sub>O), 7.02 (1H, br. s, exchangeable with D<sub>2</sub>O) and 7.31 (15H, s). [Mass spectrum: +ve ion (3NOBA, Na<sup>+</sup>)  $\text{MNa}^+$  (774)].

10

(h) Sodium (6R,7R)-7-[2-(2-Aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido-3-[(RS)-tetrahydrofuran-3-yl]ceph-3-em-4-carboxylate

15 t-Butyl (6R,7R)-7-[2-(Z)-methoxyimino-2-(2-trityl-aminothiazol-4-yl)acetamido]-3-[(RS)-tetrahydrofuran-3-yl]ceph-3-em-4-carboxylate (0.859g) was deprotected in 10% 1M hydrochloric acid in formic acid (11.4ml) as described in Example 1(h). After work up the pH of the solution was 20 adjusted to 8 with aqueous sodium hydrogen carbonate, and the product purified by column chromatography on HP20SS eluting with 1, 2, 4 and 6% THF/water. The fractions containing the product by h.p.l.c., were combined, concentrated and freeze-dried to give the title compound as 25 an amorphous white solid (0.4g, 74%);  $\nu_{\text{max}}$  (KBr) 1757, 1670, 1596 and 1532cm<sup>-1</sup>;  $\delta_{\text{H}}$  ((CD<sub>3</sub>)<sub>2</sub>SO) 1.61-2.08 (3H, m's), 3.15 and 3.37 with 3.18 and 3.37 (2H, 2ABq's, J 16.6 and 16.7Hz), 3.45-3.66 (2H, m), 3.76-3.95 (5H, m overlapping s at 3.84), 4.96 and 4.97 (1H, 2d's, J 4.3 and 4.6Hz), 5.46-5.54 (1H, m), 6.75 and 6.76 (1H, 2s's), 7.25 (2H, br s, exchangeable with D<sub>2</sub>O). [Mass spectrum: +ve ion (thioglycerol)  $\text{M}^+$  (476),  $\text{MNa}^+$  (498)].

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Example 14

4-Methoxybenzyl (6R,7R)-7-Amino-3-((R)-tetrahydrofuran-2-yl)ceph-3-em-4-carboxylate

5

(a) (R)-2-Bromoacetyltetrahydrofuran

(R)-2-Tetrahydrofuroic acid (2.739g, EPA 0382 506) was converted to it's acid chloride with oxalyl chloride (9g, 10 6.18ml) as previously described in Example 1(a). This was dissolved in dichloromethane, cooled in ice/water and saturated with excess diazomethane, bubbled through the solution in a stream of argon. 48% Aqueous hydrogen bromide (4.41ml) was then added and the reaction mixture vigorously 15 stirred. After 10min. the solution was washed with brine, dried and concentrated. Flash chromatography eluting with 5% then 10% ethyl acetate/hexane afforded the title compound as a pale yellow oil (3.519g, 77%);  $[\alpha]_D + 60.9$  (C 1.01 CHCl<sub>3</sub>).

20

(b) 4-Methoxybenzyl (RS)-2-Hydroxy-2-[(3R,4R)-3-phenyl-acetamido-4-(R)-tetrahydrofuran-2-ylcarbonylmethylthio)-azetidin-2-on-1-yl]acetate

25 4-Methoxybenzyl (RS)-2-hydroxy-2-[(1R,5R)-3-benzyl-4-thia-2,6-diazabicyclo[3.2.0]hept-2-en-7-on-6-yl]acetate (4.103g) in dichloromethane (15ml) and acetone (15ml) was ring opened with 4-toluenesulphonic acid hydrate (3.33g) in water (8ml) and coupled to (R)-2-bromoacetyltetrahydrofuran (2.11g) in 30 acetone (20ml) with potassium carbonate (0.687g) as described in Example 6(b) for the diastereoisomeric mixture. After purification by flash chromatography, the title compound was obtained as a yellow gum (2.618g, 49%);  $[\alpha]_D -10.7$  (c 1.00 CHCl<sub>3</sub>).

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(c) 4-Methoxybenzyl 2-[(3R,4R)-3-Phenylacetamido-4-[(R)-tetrahydrofuran-2-ylcarbonylmethylthio]azetidin-2-on-1-yl]2-tri-n-butylphosphoranylideneacetate

5

4-Methoxybenzyl (RS)-2-hydroxy-2-[(3R,4R)-3-phenylacetamido-4-[(R)-tetrahydrofuran-2-ylcarbonylmethylthio]azetidin-2-on-1-ylacetate (2.558g) was converted to the title compound with thionyl chloride (0.842g, 0.51ml), 2,6-lutidine (0.757g, 0.82ml) and tri-n-butylphosphine (2.1g, 2.58ml) as described for the diastereoisomeric mixture in Example 6(b). The product was obtained as a brown gum (2.16g, 63%).

(d) 4-Methoxybenzyl (6R,7R)-7-Phenylacetamido-3-[(R)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate

The phosphorane (2.16g) prepared in Example 14(c), in toluene (50ml) was heated under reflux for 8h. Removal of solvent and chromatography afforded the title compound as a yellow solid (1.008g, 67%).

(e) 4-Methoxybenzyl (6R,7R)-7-Amino-3-[(R)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate

25 The cephem (0.98g), prepared in Example 14(d) was treated with phosphorus pentachloride (0.523g) in dichloromethane (13.1ml) and N-methylmorpholine (0.429g, 0.466ml), then methanol (10mls) and water (10ml) as described for the diastereoisomeric mixture in Example 6(e). After work up 30 and purification by crystallisation from toluene, the title compound was obtained as a colourless solid (0.252g, 33%); m.p. 130-132°C;  $[\alpha]_D +11.5$  (c 1.00 CHCl<sub>3</sub>); <sup>1</sup>H n.m.r. was shown to be identical to that obtained for (R)-isomer prepared in Example 6(e).

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Example 15

4-Methoxybenzyl (6R, 7R)-7-Amino-3-((S)-tetrahydrofuran-2-yl)ceph-3-em-4-carboxylate

5

(a) (S)-2-Bromoacetyl tetrahydrofuran

(S)-2-Tetrahydrofuroic acid (5.94g) was converted to it's acid chloride with oxalyl chloride (13.00g). This was then 10 converted to the title compound with diazomethane and then 48% aqueous hydrogen bromide (9.58ml) as described in Example 14(a). After isolation, the product was obtained as pale yellow oil (8.78g, 89%);  $[\alpha]_D$  -62.8 (c 1.00,  $\text{CHCl}_3$ ).

15 (b) 4-Methoxybenzyl (RS)-2-Hydroxy-2-[(3R, 4R)-3-phenylacetamido-4-((S)-tetrahydrofuran-2-ylcarbonylmethylthio)-azetidin-2-on-1-yl]acetate

4-Methoxybenzyl (RS)-2-hydroxy-2-[(1R, 5R)-3-benzyl-4-thia-20 2, 6-diazabicyclo[3.2.0]hept-2-en-7-on-6-yl]acetate (15.09g) in 50% acetone/dichloromethane (100ml) was cleaved with 4-toluenesulphonic acid (12.25g) in water (25ml). This product was reacted with the crude bromide from Example 15(a) (8.78g) in acetone (40ml) in the presence of potassium 25 carbonate (2.53g) as described in Example 14(b). The title compound was obtained as a yellow foam (12.366g, 62%).

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(c) 4-Methoxybenzyl 2-[(3R,4R)-3-Phenylacetamido-4-[(S)-tetrahydrofuran-2-ylcarbonylmethylthio]azetidin-2-on-1-yl]2-tri-n-butylphosphoranylideneacetate

5 As for Example 14(c) the alcohol from 15(b) (12.366g) was converted to the title compound with thionyl chloride (2.47ml) and 2,6-lutidine (3.99ml) followed by tri-n-butylphosphine (12.55ml). After purification the phosphorane was obtained as a brown gum (10g, 60%).

10

(d) 4-Methoxybenzyl (6R,7R)-7-Phenylacetamido-3-[(S)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate

15 As for Example 14(d) the phosphorane from Example 15(c), (10g) was cyclized in refluxing toluene (200mls). After isolation, the title compound was obtained as a yellow foam (5.452g, 78%).

20 (e) 4-Methoxybenzyl (6R,7R)-7-Amino-3-[(S)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate

25 As for Example 14(e) the cephem from 15(d), (5.452g) was treated with phosphorus pentachloride (2.96g) and N-methylmorpholine (2.9ml) in dichloromethane (125ml), followed by treatment with methanol (50ml) then water (50ml). After adjusting the pH to 7 with 0.880 ammonium hydroxide and purification, the title compound was obtained as a pale yellow foam (2.803g, 67%); <sup>1</sup>H n.m.r. was shown to be identical to that obtained for the S-isomer prepared in 30 Example 6(c).

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Example 16

Acetoxyethyl (6R, 7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(S)-tetrahydrofuran-2-yl]ceph-3-5 em-4-carboxylate

(a) Acetoxyethyl (6R, 7R)-7-phenylacetamido-3-(tetrahydrofuran-2-yl)ceph-3-em-4-carboxylate

10 To a solution of (6R, 7R)-7-phenylacetamido-3-(tetrahydrofuran-2-yl)ceph-3-em-4-carboxylic acid (0.303g, 0.78mmol) (obtained in Example 12) in N-methylpyrrolidinone (5ml) was added potassium carbonate (0.37g, 2.66mmol). Bromomethyl acetate (0.30g, 1.95mmol) was added dropwise to the mixture 15 over 1h. The mixture was stirred for a further 1h, then ethyl acetate and water were added. The organic phase was washed with water, brine, dried ( $MgSO_4$ ) and evaporated. The residue was purified by chromatography to give the title compound as a mixture of diastereomers (0.198g, 56%); major 20 diastereomer (S);  $\delta_H$  ( $CDCl_3$ ) 1.59 (1H, m), 1.95 (2H, m), 2.12 (3H, s), 2.38 (1H, m), 3.28 (1H, d,  $J$  18.9Hz), 3.59 (1H, d), 3.65 (2H, ABq,  $J$  16.4Hz), 3.88 (2H, m), 4.89 (1H, dd,  $J$  9.0, 6.7Hz), 4.93 (1H, d,  $J$  4.9Hz), 5.84 (3H, m), 6.01 (1H, d, 9.1Hz) and 7.34 (5H, m).

25

(b) Acetoxyethyl (6R, 7R)-7-amino-3-[(S)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate

As for Example 12(b), the cephem from Example 16(a), 30 (0.196g) was treated with phosphorus pentachloride (132mg) and N-methylmorpholine (94 $\mu$ l) in dichloromethane (7ml), followed by treatment with methanol (0.85ml) then water (1.15ml). After adjusting the pH to 7 with 1M aq. ammonia and work up, the diastereomers were separated by flash 35 chromatography to give the (S)-isomer (54.3mg, 37%);  $\delta_H$  ( $CDCl_3$ ) 1.66 (1H, m), 1.97 (2H, m), 2.13 (3H, s), 2.40 (1H,

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m), 3.56 (2H, ABq, J 17.6Hz), 3.91 (2H, m), 5.03 (3H, m) and 5.84 (2H, m).

(c) Acetoxymethyl (6R,7R)-7-[(Z)-2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(S)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate

As for Example 12(c), 2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetic acid (35mg) was treated with N,N-diisopropylethylamine (34 and 27 $\mu$ l) and methanesulphonyl chloride (15 $\mu$ l) in DMF (1ml) and then added to the amino compound (53mg) obtained in Example 16(b) in DMF (1ml). After work up and chromatography the title compound (60mg, 74%) was obtained as a foam;  $\nu_{\text{max}}$  (KBr) 3330, 1774, and 1676 $\text{cm}^{-1}$ ;  $\delta_{\text{H}}$  ( $\text{CDCl}_3$ ) 1.64 (1H, m), 1.99 (2H, m), 2.14 (3H, s), 2.41 (1H, m), 3.38 and 3.67 (2H, ABq, J 18.9Hz), 3.90 (2H, m), 4.11 (3H, s), 4.95 (1H, dd, J 9.0, 6.8Hz), 5.07 (1H, d, J 4.8Hz), 5.86 (2H, m), 5.99 (1H, dd, J 8.9, 4.8Hz), 6.08 (2H, brs), 7.00 (1H, s) and 7.49 (1H, d, J 8.8Hz). [Mass spectrum: +ve ion (ammonia) 526 ( $\text{MH}^+$ )].

Example 17

Sodium (6R,7R)-7-[2-(2-Aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-(5-methoxymethyltetrahydrofuran-2-yl)ceph-3-em-4-carboxylate

(a) (2RS,5SR)-5-Methoxymethyltetrahydrofuran-2-carboxylic acid

A solution of 5-methoxymethylfuran-2-carboxylic acid (3.10g) in ethyl acetate (40ml) was hydrogenated over 5% rhodium on carbon (200mg) until hydrogen uptake ceased. The catalyst was filtered off and washed with ethyl acetate. The combined filtrates were evaporated to give the title compound as a colourless oil (3.26g);  $\nu_{\text{max}}$  (film) 1750 $\text{cm}^{-1}$ ;  $\delta_{\text{H}}$  ( $\text{CDCl}_3$ ) 1.75-2.1 (2H,m), 2.1-2.6(2H,m,), 3.45 (3H,s),

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3.47 (1H,dd, J 3 and 10 Hz), 3.74 (1H,dd, J 4 and 10Hz),  
4.15-4.43 (1H,m) and 4.43-4.63 (1H,m).

5 (b) (2RS,5SR)-2-Chloroacetyl-5-methoxymethyltetrahydrofuran

A solution of (2RS,5SR)-5-methoxymethyltetrahydrofuran-2-carboxylic acid (3.1g) in dichloromethane (50ml) was treated with oxalyl chloride (2.68ml) and dimethylformamide (1 drop). The mixture was stirred for 1h and heated to 10 reflux for 10 mins. The solvent was evaporated and then dichloromethane was evaporated from the residue twice. The product was dissolved in dichloromethane (100ml) and the solution cooled in an ice bath. Diazomethane was then passed into the solution as described in Example 14(a).  
15 When the addition was complete the mixture was stirred at 0°C for 0.5h and then hydrogen chloride gas was passed into the solution until all the diazoketone had been consumed. The solution was washed with brine, dried over magnesium sulphate and evaporated. The title compound (2.44g) was 20 isolated by column chromatography using gradient elution (silica gel, 4:1 going to 1:1 hexane : ethyl acetate);  $\nu_{\text{max}}$  (film) 1740cm<sup>-1</sup>;  $\delta_{\text{H}}$  (CDCl<sub>3</sub>) 1.6-2.35 (4H,m), 3.30-3.75 (2H,m), 3.37 (3H,s) and 4.05-4.75 (4H,m).

25 (c) (3R,4R)-4-[(2RS,5SR)-5-Methoxymethyltetrahydrofuran-2-ylcarbonylmethylthio]-3-phenoxyacetamidoazetidin-2-one

Potassium carbonate (1.0g) was added to a stirred mixture of (3R,4R)-4-mercaptop-3-phenoxyacetamidoazetidin-2-one (1.07g) 30 and (2RS,5SR) 2-chloroacetyl-5-methoxymethyltetrahydrofuran (0.869g) in dimethylformamide (15ml). The mixture was stirred at room temperature for 1.5h and then partitioned between ethyl acetate and water. The organic phase was washed twice with water, then brine, dried over magnesium 35 sulphate and evaporated. The product (0.987g) was isolated by column chromatography of the residue (silica gel, ethyl

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acetate as eluent);  $\nu_{\text{max}}$  (CHCl<sub>3</sub>) 3411, 3308, 1779 and 1689 cm<sup>-1</sup>;  $\delta_{\text{H}}$  (CDCl<sub>3</sub>) 1.63-1.77 (1H, m), 1.88-2.23 (3H, m), 3.30-3.62 (6H, m), 3.65-3.78 (1H, m), 4.15-4.30, (1H, m), 4.42-4.51 (1H, m), 4.57 (2H, s), 5.04 (1H, d, J 4.0Hz), 5.60 (1H, dd, J 4.35, 9.09Hz) 6.90-7.08 (4H, m), 7.28-7.49 (2H, m) and 7.49 (1H, t, J 8.16Hz).

(d) 4-Methoxybenzyl (2RS)-2-Hydroxy-2-[(3R,4R)-4-[(2RS,5SR)-5-methoxymethyltetrahydrofuran-2-ylcarbonyl-methylthio]-3-phenoxyacetamidoazetidin-2-on-1-yl]-acetate

A solution of 4-methoxybenzyl glyoxylate (1.82g) in dichloroethane (30ml) was heated at reflux using a Dean and Stark water separator for 1h. The solution was then cooled to room temperature and then (3R,4R)-4-[(2RS,5SR)-5-methoxymethyltetrahydrofuran-2-ylcarbonylmethylthio]-3-phenoxyacetamidoazetidin-2-one (2.94g) in dichloroethane (20ml) was added followed by triethylamine (0.1ml). The mixture was stirred at room temperature for 1h and then the solvents were removed on a rotary evaporator. The product was isolated as a mixture of isomers (3.23g) by column chromatography of the residue (silica gel, ethyl acetate as eluent);  $\nu_{\text{max}}$  (CHCl<sub>3</sub>) 3411, 1780, 1745 and 1691cm<sup>-1</sup>.

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(e) 4-Methoxybenzyl 2-[(3R,4R)-4-[(2RS,5SR)-5-methoxymethyltetrahydrofuran-2-ylcarbonylmethylthio]-3-phenoxyacetamidoazetidin-2-on-1-yl]-2-tri-n-butyl-phosphoranylideneacetate.

30

2,6-Lutidine (0.95ml) was added to a stirred solution of 4-methoxybenzyl (2RS)-2-hydroxy-2-[(3R,4R)-4-[(2RS,5SR)-5-methoxymethyltetrahydrofuran-2-ylcarbonylmethylthio]-3-phenoxyacetamidoazetidin-2-on-1-yl]acetate in tetrahydrofuran (24ml). A solution of thionyl chloride (0.59ml) in tetrahydrofuran (4ml) was then added at <-20°C

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and the mixture was stirred for 2h. The solution was filtered and evaporated and the residue was dissolved in toluene and evaporated again. The crude product was dissolved in dioxan under argon and tri-n-butylphosphine 5 (3.0ml) was added. The mixture was stirred at room temperature for 0.5h and then diluted with ethyl acetate and washed with sodium bicarbonate solution, water and brine. The solution was dried over magnesium sulphate and evaporated. The title compound (4.25g) was isolated by 10 column chromatography of the residue using gradient elution (silica gel 1:1 hexane : ethyl acetate going to neat ethyl acetate);  $\nu_{\text{max}}$  (CHCl<sub>3</sub>) 3421, 1761, 1688 and 1612 cm<sup>-1</sup>.

(f) 4-Methoxybenzyl (6R,7R)-3-[(2RS,5SR)-5-methoxymethyl-tetrahydrofuran-2-yl]-7-phenoxyacetamidoceph-3-em-4-carboxylate.

A solution of 4-methoxybenzyl 2-[(3R,4R)-4-[(2RS,5SR)-5-methoxymethyltetrahydrofuran-2-ylcarbonylmethylthio]-3-20 phenoxyacetamidoazetidin-2-on-1-yl]-2-tri-n-butylphosphoranylideneacetate (4.25g) and benzoic acid (20mg) in toluene (100ml) was heated to reflux for 10h. The mixture was cooled and the solvent evaporated. The product (1.93g) was isolated by column chromatography of the residue 25 using gradient elution (silica gel 1:1 hexane : ethyl acetate going to neat ethyl acetate);  $\nu_{\text{max}}$  (CHCl<sub>3</sub>) 3409, 1784, 1722 and 1695cm<sup>-1</sup>.

(g) 4-Methoxybenzyl (6R,7R)-7-Amino-3-(5-methoxymethyl-tetrahydrofuran-2-yl)ceph-3-em-4-carboxylate.

A solution of 4-methoxybenzyl (6R,7R)-3-[(2RS,5SR)-5-methoxymethyltetrahydrofuran-2-yl]-7-phenoxyacetamidoceph-3-em-4-carboxylate (1.93g) in dichloromethane (25ml) was

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cooled to -15 to -20°C, N-methylmorpholine (0.75ml) was added followed by a solution of phosphorus pentachloride in dichloromethane (26.5ml of a solution containing 40mg.ml<sup>-1</sup>). The mixture was stirred at the same temperature for 0.5h and 5 then methanol (6.8ml) was added and the mixture stirred at room temperature for 0.5h. Water (10ml) was then added and the mixture vigorously stirred for 0.5h. The dichloromethane was then removed on a rotary evaporator and the residue was partitioned between ether and water. The 10 aqueous phase was stirred with ethyl acetate and the pH was adjusted to 6.2 with 1N aqueous ammonia. The organic phase was washed with water and brine, dried over magnesium sulphate and evaporated. The products were separated by column chromatography of the residue using gradient elution 15 (silica gel, 1:1 hexane : ethyl acetate going to neat ethyl acetate). The following were obtained in order of elution : 4-methoxybenzyl (6R, 7R)-7-amino-3-[(2S, 5R)-5-methoxy- methylethyltetrahydrofuran-2-yl]ceph-3-em-4-carboxylate (388mg);  $\nu_{\text{max}}$  (CHCl<sub>3</sub>) 3410, 1776 and 1725cm<sup>-1</sup>;  $\delta_{\text{H}}$  (CDCl<sub>3</sub>) 1.59-1.78 20 (2H, m), 1.93-2.08 (1H, m), 2.18-2.32 (1H, m), 2.54 (2H, br s), 3.33-3.54 (3H, m), 3.38 (3H, s), 3.80 (3H, s), 4.00-4.11 (1H, m), 4.76 (1H, d, J 4.99Hz), 4.90 (1H, d, J 4.97Hz) 4.96 (1H, t, J 8.23Hz) 5.17 (2H, s), 6.88 (2H, d, J 8.60Hz) and 7.33 (2H, d, J 8.61Hz). [Mass spectrum: M<sup>+</sup> (434)]; 4- 25 methoxybenzyl (6R, 7R)-7-amino-3-[(2R, 5S)-5- methoxymethylethyltetrahydrofuran-2-yl]ceph-3-em-4-carboxylate (305mg);  $\nu_{\text{max}}$  (CHCl<sub>3</sub>) 3409, 1776 and 1725cm<sup>-1</sup>;  $\delta_{\text{H}}$  (CDCl<sub>3</sub>) 1.60-1.81 (2H, m), 1.85-2.01 (2H, m), 3.30-3.50 (2H, m), 3.38 (3H, s), 3.44 (1H, d, J 17.78Hz), 3.69 (1H, d, J 30 17.75Hz), 3.80 (3H, s), 4.00-4.17 (1H, m), 4.70 (1H, d, J 4.92Hz), 4.93 (1H, d, J 4.95Hz), 5.10-5.20 (1H, m), 5.18 (1H, d, J 11.88Hz), 5.24 (1H, d, J 11.89Hz), 6.88 (1H, d, J 8.65Hz) and 7.35 (1H, d, J 8.64Hz). [Mass spectrum: M<sup>+</sup> (434)].

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(h) 4-Methoxybenzyl (6R,7R)-7-[2-(2-Aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(2R,5S)-5-methoxymethyltetrahydrofuran-2-yl]ceph-3-em-4-carboxylate

5 A stirred solution of 2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetic acid (155mg) and N,N-diisopropyl-ethylamine (134 $\mu$ l) in dimethylformamide (3ml) was cooled to -30° to -40°C and methanesulphonyl chloride (60 $\mu$ l) was added. The mixture was stirred at the same temperature for 10 0.5h and then a solution of 4-methoxybenzyl (6R,7R)-7-amino-[(2R,5S)-5-methoxymethyltetrahydrofuran-2-yl]ceph-3-em-4-carboxylate (304mg) in dimethylformamide (3ml) was added followed by pyridine (60 $\mu$ l). The mixture was then stirred at 0°C for 1.5h, and then partitioned between ethyl acetate 15 and aqueous citric acid solution. The organic phase was washed three times with water, then with brine, dried over magnesium sulphate and evaporated. The title compound (115mg) was isolated by column chromatography of the residue using gradient elution (silica gel 1:1 hexane : ethyl acetate going to neat ethyl acetate),  $\nu_{\text{max}}$  (CHCl<sub>3</sub>) 3489, 3397, 3330, 1779, 1723, 1681cm<sup>-1</sup>;  $\delta_{\text{H}}$  (CDCl<sub>3</sub>) 1.60-1.80 (2H,m), 1.78-2.05 (2H,m), 3.30-3.53 (3H,m), 3.37 (3H,s), 3.70 (1H,d, J 17.87Hz), 3.81 (3H,s), 4.08 (3H,s), 5.05 (1H,d, J 4.79Hz), 5.18 (1H,d, J 11.81Hz), 5.24 (1H,d, J 25 11.62Hz), 5.90 (1H,dd, J 4.75 and 8.89Hz), 6.90 (1H,d, J 9.56Hz), 6.91 (1H,s), 7.34 (1H,d, J 8.67Hz) and 7.67 (1H,d, J 8.88Hz).

(i) Sodium (6R,7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(2SR,5S)-5-methoxymethyltetrahydrofuran-2-yl]ceph-3-em-4-carboxylate.

Concentrated hydrochloric acid (0.15ml) was added to a stirred solution of 4-methoxybenzyl (6R,7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(2R,5S)-5-methoxymethyltetrahydrofuran-2-yl]ceph-3-em-4-carboxylate (115mg) in 95% formic acid (4ml). The

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mixture was stirred at room temperature for 1.5h and then the solvents were removed on a rotary evaporator, and then toluene was evaporated from the residue twice. The residue was stirred with water and toluene and the pH of the aqueous phase was adjusted to 6.2 with aqueous sodium bicarbonate solution. The aqueous phase was separated and evaporated and the title compound (36mg) was obtained as a mixture of isomers by column chromatography of the residue (HP20SS water with increasing proportions of acetone as eluent).

10 Fractions containing product were combined, evaporated, and the residue dissolved in water (5ml) and freeze-dried;  $\nu_{max}$  (KBr) 1762, 1671 and 1602  $\text{cm}^{-1}$ ;  $\delta_{\text{H}}$  [(CD<sub>3</sub>)<sub>2</sub>SO] 1.4-2.15 (4H, m), 3.14-3.48 (4H, m), 3.24 and 3.27 (3H, 2s), 3.83 (3H, s), 3.87-3.98 and 4.03-4.18 (1H, m), 4.96 (1H, d,  $J$  15 4.66Hz), 5.00 and 5.22 (1H, 2t,  $J$  7.47Hz), 5.46-5.57 (1H, m), 6.74 and 6.75 (1H, 2s), 7.25 (2H, s) and 9.49 and 9.53 (1H, 2d,  $J$  8.12Hz)

Example 18

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Sodium (6R,7R)-7-[2-(2-Aminothiazol-4-yl)-(Z)-pent-2-enamido-3-[(S)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate.

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(a) 4-Methoxybenzyl (6R,7R)-7-[2-(2-Aminothiazol-4-yl)-(Z)-pent-2-enamido-3-[(S)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate.

Mesyl chloride (70 $\mu$ l) was added to 2-(2-aminothiazol-4-yl)-  
30 (Z)-pent-2-enoic acid (178mg) and N,N-diisopropylethylamine (160 $\mu$ l) in DMF (5ml) and dichloromethane (5ml) at -20°C. The reaction mixture was stirred at -20°C for 1 hour then added to an icecold solution of 4-methoxybenzyl (6R,7R)-7-amino-3-[(S)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate (370mg) and N,N-diisopropylethylamine (160 $\mu$ l) in dichloromethane (5ml). Stirred for 1 hour, concentrated and flash chromatographed on silica gel eluting with 30, 50, 60 and 70% ethyl acetate in hexane to give the title compound

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(90mg);  $\nu_{\text{max}}$  (CHCl<sub>3</sub>) 1782, 1720, 1674, 1614, 1516, 1134 and 1107cm<sup>-1</sup>;  $\delta_{\text{H}}$  (CDCl<sub>3</sub>, 250MHz) 1.12 (3H, t, J 7.5Hz) 1.50-2.45 (6H, m), 3.30-3.95 (7H, m), 4.85-5.05 (2H, m), 5.18 (2H, s), 5.85-5.95 (1H, m), 6.44 (1H, s), 6.52 (1H, t, J 7.8Hz), 6.90 and 7.32 (4H, ABq, J 8.6Hz) and 7.43 (1H, d, J 8Hz). [Mass spectrum: +ve ion (3-nitrobenzyl alcohol, sodium acetate)  $\text{MNa}^+$  (689)].

(b) Sodium (6R, 7R)-7-[2-(2-Aminothiazol-4-yl)-(Z)-pent-2-enamido]-3-[(S)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate

4-Methoxybenzyl (6R, 7R)-7-[2-(2-aminothiazol-4-yl)-(Z)-pent-2-enamido]-3-[(S)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate (80mg) in dichloromethane (2ml) was added 15 dropwise to a mixture of aluminium chloride (47mg) and anisole (1.03ml) in dichloromethane (2ml) at -50°C under argon. The mixture was stirred for 15 minutes at -40°C and 0.5M trisodium citrate (3.42ml) added, stirred at room temperature for 15 minutes then diluted with dichloromethane 20 (10ml) and water (10ml). The aqueous layer was collected, washed with dichloromethane and chromatographed on HP20SS eluting with 0, 1, 2, 5 and 10% acetone in water. Fractions containing the product, h.p.l.c analysis, were combined, concentrated and freeze-dried to give the title compound 25 (22mg);  $\nu_{\text{max}}$  (KBr) 3407, 1757, 1609, 1527, 1375, 1338 and 1041 cm<sup>-1</sup>;  $\delta_{\text{H}}$  (D<sub>2</sub>O, 250MHz) 1.03 (3H, t, J 7.5Hz), 1.65-2.30 (6H, m), 3.32 and 3.51 (2H, ABq, J 7.7Hz), 3.70-3.95 (2H, m), 4.65-4.80 (1H, m), 5.20 (1H, d, J 4.7Hz), 5.74 (1H, d, J 4.7Hz), 6.33 (1H, t, J 8Hz) and 6.47 (1H, s). [Mass spectrum +ve ion 30 (thioglycerol)  $\text{MH}^+$  (473)].

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Example 19

Sodium (6R, 7R)-7-[2-(2-Aminothiadiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(S)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate

(a) 4-Methoxybenzyl (6R, 7R)-7-[2-(Z)-Methoxyimino-2-(2-tritylaminothiadiazol-4-yl)acetamido]-3-[(S)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate.

10 Mesyl chloride (65 $\mu$ l) was added to 2-(Z)-methoxyimino-2-(2-tritylaminothiadiazol-4-yl)acetic acid (370mg) and N,N-diisopropylethylamine (146 $\mu$ l) in dichloromethane (5ml) at -20°C. The reaction mixture was stirred at -20°C for 1 hour 15 then added to an ice cold solution of 4-methoxybenzyl (6R, 7R)-7-amino-3-[(S)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate (335mg) and pyridine (70 $\mu$ l) in dichloromethane (5ml). The reaction was stirred for 1 hour, concentrated and flash chromatographed on silica gel eluting with 20 30, 50, 60 and 70% ethyl acetate in hexane to afford the title compound as a foam (300mg);  $\nu_{\text{max}}$  (CHCl<sub>3</sub>) 3398, 1784, 1724, 1691, 1516, 1134 and 1107 cm<sup>-1</sup>;  $\delta_{\text{H}}$  (CDCl<sub>3</sub>, 250MHz) 1.55-1.75 (1H, m), 1.80-2.05 (2H, m), 2.25-2.45 (1H, m), 3.30 and 3.61 (2H, ABq, J 18.3Hz), 3.75-4.00 (2H, m), 3.81 (3H, m), 4.16 (3H, m), 4.85-5.00 (1H, m), 5.00 (1H, d, J 4.8Hz), 5.17 (2H, s), 5.92 (1H, dd, J 4.8Hz), 6.72 (1H, d, J 7.8Hz) and 6.88 and 7.30 (19H, m). [Mass spectrum: +ve ion (3-nitrobenzyl alcohol, sodium acetate)  $\text{MNa}^+$  (839)].

30 (b) Sodium (6R, 7R)-7-[2-(2-Aminothiadiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(S)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate

Trifluoroacetic acid (5ml) was added to 4-methoxybenzyl (6R, 7R)-7-[2-(Z)-methoxyimino-2-(2-tritylaminothiadiazol-4-

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yl)acetamido]-3-[(*S*)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate (100mg) and anisole (1ml) in dichloromethane (5ml) at room temperature and stirred for 1 hour. The mixture was evaporated and re-evaporated from toluene-5 (10ml). The residue was dissolved in 1% sodium hydrogen carbonate solution (1ml), washed with ether and chromatographed on HP20SS eluting with 0, 0.5 and 1% acetone in water. Fractions containing the product, h.p.l.c analysis, were combined, concentrated and freeze dried to 10 give the title compound (35mg);  $\nu_{\text{max}}$  (KBr) 3381, 1758, 1669, 1602, 1527, 1399 and 1042cm<sup>-1</sup>;  $\delta_{\text{H}}$  (D<sub>2</sub>O) 1.64-2.25 (4H, m), 3.30 and 3.49 (2H, ABq, J 17.8Hz), 3.70-3.95 (2H, m), 4.03 (3H, s), 4.65-4.75 (1H, m), 5.19 (1H, d, J 4.7Hz) and 5.77 (1H, d, J 4.7Hz). [Mass spectrum: +ve ion (thioglycerol) 15  $\text{MH}^+$  (477)].

Example 20

(*RS*)-1-Acetoxyethyl (6*R*,7*R*)-7-[2-(2-Aminothiazol-4-yl)-2-20 (*Z*)-methoxyiminoacetamido]-3-[(*S*)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate.

A solution of (*RS*)-1-acetoxyethylbromide (267mg) in 1-methyl-2-pyrrolidinone (2ml) was added dropwise, over 1 25 hour, to an ice cold mixture of sodium (6*R*,7*R*)-7-[2-(2-aminothiazol-4-yl)-2-(*Z*)-methoxyiminoacetamido]-3-[(*S*)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate (190mg) and potassium carbonate (110mg) in 1-methyl-2-pyrrolidinone (1ml). After 15 minutes the mixture was diluted with ethyl 30 acetate, washed with water, brine, dried (MgSO<sub>4</sub>), concentrated and flash chromatographed on silica gel eluting with 50, 70, 80 and 90% ethyl acetate in hexane to give the title compound (172mg);  $\nu_{\text{max}}$  (CHCl<sub>3</sub>) 3019, 2929, 1786, 1683, 1520, 1376 and 1135cm<sup>-1</sup>;  $\delta_{\text{H}}$  (CDCl<sub>3</sub>, 250MHz) 1.45-1.75 (4H, m), 1.90-2.10 (2H, m), 2.09 and 2.10 (together 3H, 2s), 2.30-2.50 (1H, m), 3.36 and 3.65 (2H, ABq, J 18.8Hz), 4.93-35 5.10 (2H, m), 5.90-6.05 (1H, m), 6.94 and 7.07 (together 5.10 (2H, m), 5.90-6.05 (1H, m), 6.94 and 7.07 (together

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1H, q, J 5.8Hz), 7.10 and 7.15 (together 1H, 2s) and 7.60 and 7.67 (together 1H, 2d, J 7.4Hz); [Mass spectrum: +ve ion (3-nitrobenzyl alcohol, sodium acetate)  $MNa^+$  (562)].

5 Example 21

(6R,7R)-7-[2-(2-Aminothiazol-4-yl)-2-(Z)-carboxymethoxy-iminoacetamido]-3-[(RS)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylic acid disodium salt.

10

(a) 4-Methoxybenzyl (6R,7R)-7-[2-(Z)-t-butoxycarbonyl-methoxyimino-2-(2-tritylaminothiazol-4-yl)acetamido]-3-[(R)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate.

15

2-[(2)-t-Butyloxycarbonylmethoxyimino]-2-(2-tritylaminothiazol-4-yl)acetic acid (179mg, 0.31mmol) in DMF (4ml) was treated at -25°C with N,N-diisopropylethylamine (52 $\mu$ l, 0.31m mol) and methanesulphonyl chloride (24 $\mu$ l, 0.31m mol) for 30min. A mixture of 4-methoxybenzyl (6R,7R)-7-amino-3-[(R)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate (119mg, 0.31mmol) [See example 6] and pyridine (26 $\mu$ l, 0.31mmol) dissolved in DMF (4ml) was added and stirring was maintained at 0°C for 1h. The reaction mixture was partitioned between ethyl acetate and dilute aqueous sodium hydrogen carbonate, 25 the organic layer was washed with aqueous citric acid then water, dried (magnesium sulphate) and evaporated to low bulk. The residue was chromatographed on silica gel eluting with ethyl acetate/hexane mixtures to give the title compound as a cream amorphous solid (190mg, 69%);  $\delta_H$  (CDCl<sub>3</sub>) 1.43 (9H, s), 1.54-1.68 (2H, m), 1.86-1.95 (1H, m), 2.02-2.12 (1H, m), 3.34 and 3.50 (2H, ABq, J 18Hz), 3.76-3.91 (2H, m), 3.81 (3H, s), 4.76 (2H, br s), 5.02 (1H, d, J 5Hz), 5.16-5.22 (1H, m), 5.78 (1H, dd, J 5, 8Hz), 6.84 (1H, s), 6.8 (2H, d, J 9Hz), 7.0 (1H, br s, exch) and 7.26-7.36 (17H, m). [Mass spectrum: +ve ion (3-nitrobenzylalcohol, sodium acetate)  $MNa^+$  (938)].

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(b) (6R,7R)-7-[2-Aminothiazol-4-yl)-2-(Z)-carboxymethoxy-iminoacetamido]-3-[(RS)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylic acid disodium salt.

5 The product of Example 21(a) (174mg, 0.19m mol) was dissolved in a mixture of trifluoroacetic acid; dichloromethane and anisole (4:4:1, 5ml) and kept at room temperature for 2h. The solution was evaporated to dryness under reduced pressure and the residue was twice washed with 10 ether. The residue solid was dissolved in water using sodium hydrogen carbonate to bring to pH7.5 then the solution was chromatographed on HP20SS eluting with water. There was some separation of isomers but most of the product was collected as a mixed fraction of (R) and (S) 15 tetrahydrofuryl isomers which was freeze dried to a white solid (42mg, 44%),  $\nu_{\text{max}}$  (KBr) 1761, 1660, (sh) 1601 and 1533  $\text{cm}^{-1}$ ;  $\delta_{\text{H}}$  ( $\text{D}_2\text{O}$ ) (major isomer) 1.69-2.18 (4H,m), 3.32 and 3.51 (2H, ABq, J 18Hz), 3.74-3.93 (2H,m), 4.52 (2H,s), 5.19 (1H, d, J 5Hz), 5.77 (1H, d, J 5Hz) and 7.01 (1H,s); 20 (minor isomer) (inter alia), 3.37 and 3.57 (ABq, J 17Hz), 5.17 (d, J 5Hz) and 5.76 (d, J 5Hz). [Mass spectrum: +ve ion (3-nitrobenzyl alcohol, sodium acetate)  $\text{MNa}^+$  (563)].

Example 22

25

Sodium (6R,7R)-7-[(R)-2-Amino-2-(4-hydroxyphenyl)acetamido]-3-[(S)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate.

(a) 4-Methoxybenzyl (6R,7R)-7-[(R)-2-t-butoxycarbonylamino-30 2-(4-hydroxyphenyl)acetamido]-3-[(R)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate

4-Methoxybenzyl (6R,7R)-7-amino-3-[(R)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate (136mg, 0.35mmol) [See example 6] 35 in THF (10ml) was stirred in an ice bath with

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dicyclohexylcarbodiimide (108mg, 0.52mmol) then (R)-2-t-butoxycarbonylamino-2-(4-hydroxyphenyl)acetic acid (139mg, 0.52mmol) in THF (3ml) was added dropwise over 2 min. The mixture was stirred at 0°C for 30 min then at room temperature for 30min. It was filtered and evaporated and the residue chromatographed on silica gel eluting with ethyl acetate/hexane mixtures. The title compound was obtained as a white solid (212mg, 95%);  $\delta_H$  (CDCl<sub>3</sub>) 1.10-2.0 (4H,m), 1.42 (9H,s), 3.18 and 3.43 (2H,ABq, J 17Hz), 3.80 (3H,s), 3.77-10 3.88 (2H,m), 4.89 (1H,d, J 5Hz), 5.10 (1H,t, J 7Hz), 5.11 (1H,d, J 5Hz), 5.19 (2H, s), 5.65 (1H,d, J 5Hz exch), 5.69 (1H,dd, J 4,9Hz), 6.72 (2H,d, J 8Hz), 6.81 (1H,d, J 9Hz exch), 6.88 (2H,d, J 9Hz), 7.10 (2H,d, J 8Hz) and 7.33 (2H,d,J 9Hz). [Mass spectrum: +ve ion (3-nitrobenzyl alcohol, sodium acetate)  $MNa^+$  (938)].

(b) Sodium (6R,7R)-7-[(R)-2-Amino-2-(4-hydroxyphenyl)-acetamido]-3-[(RS)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate

20 The product of Example 22(a) (42mg, 0.66mmol) was treated as in Example 21(b). The final chromatography on HP20SS yielded two fractions. The first fraction to be eluted was the pure (S)-tetrahydrofuran-2-yl isomer (53mg, 19%) as a white freeze dried solid;  $\nu_{max}$  (KBr) 1762, 1690 and 1600cm<sup>-1</sup>;  $\delta_H$  (D<sub>2</sub>O) 1.62-1.74 (1H,m), 1.87-1.98 (2H,m), 2.15-2.05 (1H,m), 3.10 and 3.39 (2H, ABq, J 18Hz), 3.72-3.90 (2H,m), 4.66 (1H,t, J 8Hz), 5.04 (1H,d, J 4.5Hz), 5.61 (1H,d, J 4.5Hz), 6.90 (1H,d, J 9HZ) and 7.31 (2H,d, J 9Hz). 30 Further elution of the column gave a mixture of diastereoisomers (84mg, 30%).

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Example 23

Sodium (1S, 6R, 7R)-7-[2-(2-Aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(S)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate-1-oxide

(a) 4-Methoxybenzyl (1S, 6R, 7R)-7-[2-(2-Aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(S)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate-1-oxide

10

4-Methoxybenzyl (6R, 7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(S)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate (see example 7) (250mg, 0.44mmol) in ethyl acetate (25ml) was stirred in an ice bath and a solution of 15 m-chloroperbenzoic acid (75mg, 0.44mmol) in ethyl acetate (5ml) was added. After 10 min the reaction mixture was washed with dilute aqueous sodium hydrogen carbonate then water followed by drying (magnesium sulphate) and evaporation under reduced pressure. The residue was chromatographed on silica gel eluting with acetone/ethyl acetate mixtures to give the title compound as a white solid (179mg, 69%);  $\nu_{\text{max}}$  (CHCl<sub>3</sub>) 1800, 1730, 1680 and 1610 cm<sup>-1</sup>;  $\delta_{\text{H}}$  (CDCl<sub>3</sub>) 1.48-1.64 (1H, m), 1.89-2.00 (2H, m), 2.33-2.47 (1H, m), 3.29 and 3.75 (2H, ABq, J 19Hz), 3.82 (3H, s), 3.84-25 3.96 (2H, m), 4.09 (3H, s), 5.06 (1H, dd, J 7, 9Hz), 5.22 (2H, s), 5.55-5.8 (1H, br s, exch), 6.16 (1H, dd, J 4.5, 10Hz), 6.91 (2H, d, J 7, 9Hz), 6.98 (1H, s), 7.35 (2H, d, J 9Hz) and 7.55-7.65 (1H, br, exch). [Mass spectrum: +ve ion (thioglycerol)  $\text{MH}^+$  (590)].

30

(b) Sodium (1S, 6R, 7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(S)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate-1-oxide

35 Anhydrous aluminium chloride (115mg, 0.86mmol) was added to a mixture of anisole (5ml) and dichloromethane (3ml) cooled

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to -20°C. After 15 mins at -20°C the mixture was cooled to -40°C and a solution of the product of Example 23(a) (1.70mg, 0.29mmol) in dichloromethane (4ml) was then added. The mixture was then stirred at -40°C for 10 min when a 0.5M aqueous solution of trisodium citrate (9ml) was added. After vigoursly stirring at room temperature the aqueous layer was separated, twice washed with dichloromethane then concentrated under reduced pressure. The residue was chromatographed on HP20SS eluting with water containing up to 2% acetonitrile. Pure fractions (as determined by HPLC) were combined and freeze dried to give the title compound as a white solid (71mg, 50%);  $\nu_{\text{max}}$  (KBr) 1775, 1669 and 1607 (br)  $\text{cm}^{-1}$ ;  $\delta_{\text{H}}$  ( $\text{D}_2\text{O}$ ) 1.54-1.70 (1H, m), 1.94-2.03 (2H, m), 2.15-2.28 (1H, m), 3.44 and 3.85 (2H, ABq,  $J$  18Hz), 3.8-4.0 (2H, m), 3.99 (3H, s), 4.86 (1H, t,  $J$  8Hz), 4.99 (1H, d,  $J$  4.5Hz), 5.95 (1H, d,  $J$  4.5Hz) and 7.01 (1H, s). [Mass spectrum: +ve ion (thioglycerol)  $\text{MH}^+$  (492)].

Example 24

20

Sodium 7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-(tetrahydrofuran-2-yl)-1-carba-1-dethiaceph-3-em-4-carboxylate

25 (a) 4-Methoxybenzyl 2-diazo-3-oxo-5-[(3SR,4RS)-3-phenylacetamidoazetidin-2-on-4-yl]pentanoate

A solution of 4-methoxybenzyl 3-oxo-5-[(3SR,4RS)-3-phenylacetamidoazetidin-2-on-4-yl]pentanoate (1.38g, 3.15mmol) [prepared by the method described for 4-nitrobenzyl 3-oxo-5-[(3SR,4RS)-3-phenoxyacetamidoazetidin-2-on-4-yl]pentanoate, C. Bodurow and M. A. Carr, Tetrahedron Lett., 1989, 30, 4801] in acetonitrile (60ml) was treated with 4-toluenesulphonyl azide (870mg, 4.42mmol) and N,N-diisopropylethylamine (822 $\mu$ l, 4.73mmol) at 0°C. After 10 min., the ice-bath was removed and stirring was continued at

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room temperature for 2h. The reaction mixture was diluted with ethyl acetate and washed with brine. After drying over  $MgSO_4$ , the solvent was evaporated in vacuo and the residue purified by chromatography on silica gel eluting with-ethyl acetate to yield the title compound (1.27g, 87%);  $\nu_{max}$  (KBr) 2134, 1775, 1717, 1654, 1513 and  $1304\text{cm}^{-1}$ ;  $\delta_H$  ( $CDCl_3$ , 250MHz) 1.59-1.70 (2H,m), 2.68-2.95 (2H,m), 3.55 and 3.65 (2H, ABq, J 15.6Hz), 3.78 (1H,m), 3.82 (3H,s), 5.19 (2H,s), 5.25 (1H, ddd, J 8.1, 4.9, 1.0Hz), 6.25 (1H, br s, exch.), 6.49 (1H, br d, J 8.1Hz, exch.), 6.90 (2H,d, J 8.7Hz) and 7.23-7.70 (7H,m). [Mass spectrum : +ve ion (3-nitrobenzyl alcohol, sodium acetate)  $MNa^+$  (487)].

(b) 4-Methoxybenzyl (6RS,7SR)-7-Phenylacetamido-3-(trifluoromethylsulphonyloxy)-1-carba-1-dethiaceph-3-em-4-carboxylate

A solution of 4-methoxybenzyl 2-diazo-3-oxo-5-[(3SR,4RS)-3-phenylacetamidoazetidin-2-on-4-yl]pentanoate (1.54g, 3.32mmol) in chloroform (40ml) was heated to reflux in the presence of a catalytic quantity of rhodium (II) acetate dimer. After heating for 45min., the reaction mixture was cooled to  $0^\circ\text{C}$  and treated sequentially with N,N-diisopropylethylamine (1.16ml, 6.66mmol) and trifluoromethanesulphonic anhydride (0.61ml, 3.65mmol). After stirring for 30 min at  $0^\circ\text{C}$ , the mixture was concentrated in vacuo. The residue was purified by chromatography on silica gel eluting with 30, then 50% ethyl acetate in hexane yielding the title compound as an orange foam (1.20g, 64%);  $\nu_{max}$  ( $CH_2Cl_2$ ) 3417, 1783, 1733, 1684, 1516 and  $1430\text{cm}^{-1}$ ;  $\delta_H$  ( $CDCl_3$ , 250MHz) 1.45 (1H,m), 2.01 (1H,m), 2.56 (2H,m), 3.58 and 3.64 (2H, ABq, J 16.1Hz), 3.80 (3H,s), 3.87 (1H,m), 5.14-5.35 (3H,m), 5.89 (1H, br d, J 6.2Hz, exch), 6.87 (2H, d, J 8.7Hz) and 7.22-7.41 (7H,m),. [Mass spectrum: +ve ion (ammonia)  $MH^+$  (569),  $MNH_4^+$  (586)].

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(c) 4-Methoxybenzyl (6RS,7SR)-7-Phenylacetamido-3-[ (RS)-tetrahydrofuran-2-yl]-1-carba-1-dethiaceph-3-em-4-carboxylate

5 A solution of 4-methoxybenzyl (6RS,7SR)-7-Phenylacetamido-3-(trifluoromethylsulphonyloxy)-1-carba-1-dethiaceph-3-em-4-carboxylate (1.13g, 199mmol) in THF (15ml) was treated with the cuprate species generated from (tetrahydrofuran-2-yl)-tri-n-butylstannane (1.97g, 5.46mmol), n-butyllithium (4.1ml 10 of a 1.6M solution in hexane, 6.56mmol) and copper (I) bromide dimethylsulphide complex (565mg, 2.75mmol) as described in Example 11(a). Following work-up, the crude reaction product was purified by chromatography on silica gel eluting with 10, 20 and 30% ethyl acetate hexane. After 15 elution of the 3-n-butylcarbacephem (340mg, 36%), the title compound was obtained as a mixture of diastereoisomers (478mg, 50%); (found: M<sup>+</sup>, 490.2096. C<sub>28</sub> H<sub>30</sub> N<sub>2</sub> O<sub>6</sub> requires M<sup>+</sup> 490.2104.);  $\nu_{\text{max}}$  (CH<sub>2</sub>Cl<sub>2</sub>) 3422, 1769, 1719, 1682, 1515 and 1389cm<sup>-1</sup>;  $\delta_{\text{H}}$  (CDCl<sub>3</sub>, 250MHz) 1.45-2.70 (8H,m), 3.58 and 20 3.67 (2H, ABq, J 16.0Hz), 3.72-3.90 (3H,m), 3.80 (3H,s), 4.93 and 5.09 (together 1H, 2dd, J 8.9, 6.8 and 7.9, 7.9Hz), 5.13-5.28 (3H,m), 5.80 and 5.85 (together 1H, 2d, J 6.6, 7.7Hz, exch.), 6.89 (2H,d, J 8.7Hz) and 7.20-7.41 (7H,m).

25 (d) 4-Methoxybenzyl (6RS,7SR)-7-amino-3-(tetrahydrofuran-2-yl)-1-carba-1-dethiaceph-3-em-4-carboxylate

A solution of 4-methoxybenzyl (6RS, 7SR)-7-phenylacetamido-3-[ (RS)-tetrahydrofuran-2-yl]-1-carba-1-dethiaceph-3-em-4-30 carboxylate (560mg, 1.14mmol) and N-methylmorpholine (250 $\mu$ l, 2.27mmol) in dichloromethane (15ml) was treated successively with phosphorus pentachloride (357mg, 1.71mmol) in dichloromethane (9ml), methanol (2.5ml) and water (5ml) as described in Example 1(f). Purification by chromatography 35 on silica gel eluting with ethyl acetate and then 5% methanol in ethyl acetate yielded 4-methoxybenzyl (6RS,7SR)-7-amino-3-[ (SR)-tetrahydrofuran-2-yl]-1-carba-1-dethiaceph-

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3-em-4-carboxylate (166mg, 39%) as a colourless foam;  $\nu_{\text{max}}$  ( $\text{CH}_2\text{Cl}_2$ ) 3401, 1761, 1716, 1613 and  $1516\text{cm}^{-1}$ ;  $\delta_{\text{H}}$  ( $\text{CDCl}_3$ , 250MHz) 1.50-1.68 (2H, m), 1.85-1.97 (2H, m), 2.12-2.32 (2H, m), 2.35-2.45 (2H, m), 2.70 (2H, br s, exch.), 3.70-3.92 (3H, m), 3.78 (3H, s), 4.58 (1H, d,  $J$  5.3Hz), 4.94 (1H, dd,  $J$  8.8, 7.0Hz), 5.16 (2H, s), 6.87 (2H, d,  $J$  8.7Hz) and 7.31 (2H, d,  $J$  8.7Hz). [Mass spectrum: +ve ion (3-nitrobenzyl alcohol, sodium acetate)  $\text{MNa}^+$  (395)]. Further elution of the column yielded the more polar diastereoisomer 4-10 methoxybenzyl (6RS, 7SR)-7-amino-3-[(RS)-tetrahydrofuran-2-yl]-1-carba-1-dethiaceph-3-em-4-carboxylate (132mg, 31%) as a pale yellow foam;  $\nu_{\text{max}}$  ( $\text{CH}_2\text{Cl}_2$ ) 3408, 1761, 1722, 1613 and  $1516\text{cm}^{-1}$ ;  $\delta_{\text{H}}$  ( $\text{CDCl}_3$ , 250MHz) 1.52-1.72 (2H, m), 1.80-2.00 (2H, m), 2.06-2.22 (2H, m), 2.50-2.78 (4H, m, 2H exch), 3.69-15 3.90 (3H, m), 3.78 (3H, s), 4.51 (1H, d,  $J$  5.3Hz), 5.06 (1H, dd,  $J$  7.8, 7.8Hz), 5.20 (2H, s), 6.87 (2H, d,  $J$  8.6Hz) and 7.34 (2H, d,  $J$  8.6Hz). [Mass spectrum : +ve ion (3-nitrobenzyl alcohol, sodium acetate)  $\text{MNa}^+$  (395)].

20 (e) 4-Methoxybenzyl (6RS, 7SR)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyaminoacetamido]-3-[(SR)-tetrahydrofuran-2-yl]-1-carba-1-dethiaceph-3-em-4-carboxylate

2-(2-Aminothiazol-4-yl)-2-(Z)-methoxyiminoacetic acid (99mg, 0.49mmol) in DMF (5ml) was treated with methansulphonyl chloride (38 $\mu$ l, 0.49mmol) and N,N-diisopropylethylamine (86 $\mu$ l, 0.49mmol) as described in Example 7(a). This was then treated successively with a solution of 4-methoxybenzyl (6RS, 7SR)-7-amino-3-[(SR)-30 tetrahydrofuran-2-yl]-1-carba-1-dethiaceph-3-em-4-carboxylate (160mg, 0.43mmol) in DMF (5ml) and pyridine (40 $\mu$ l, 0.49mmol). After work-up, the product was purified by chromatography on silica gel eluting with ethyl acetate to yield the title compound (169mg, 71%);  $\nu_{\text{max}}$  (KBr) 3313, 35 1763, 1717, 1676, 1612 and  $1514\text{cm}^{-1}$ ;  $\delta_{\text{H}}$  ( $\text{CDCl}_3$ , 250MHz) 1.48-1.62 (2H, m), 1.83-1.98 (2H, m), 2.10-2.49 (6H, m, 2H exch.), 3.78-3.98 (3H, m), 3.79 (3H, s), 4.08 (3H, s), 4.98 (1H, dd,  $J$  8.8, 6.9Hz), 5.13 and 5.20 (2H, ABq,  $J$  12.2Hz),

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5.48 (1H, dd, J 7.0, 5.0Hz), 6.89 (2H, d, J 8.6Hz), 7.00 (1H, s), 7.35 (2H, d, J 8.6Hz) and 7.82 (1H, br s, exch.). [Mass spectrum: +ve ion (3-nitrobenzylalcohol, sodium acetate)  $\text{MH}^+$  (556)  $\text{MNa}^+$  (578)].

5

(f) 4-methoxybenzyl (6RS,7SR)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(RS)-tetrahydrofuran-2-yl]-1-carba-1-dethiaceph-3-em-4-carboxylate

10 2-(2-Aminothiazol-4-yl)-2-(Z)-methoxyiminoacetic acid (74mg, 0.37mmol) in DMF (5ml) was treated with methanesulphonyl chloride (29 $\mu$ l, 0.37mmol) and N,N-diisopropylethylamine (64 $\mu$ l, 0.37mmol) as described in Example 7(a). This was then treated successively with a solution of 4-methoxybenzyl 15 (6RS,7SR)-7-amino-3-[(RS)-tetrahydrofuran-2-yl]-1-carba-1-dethiaceph-3-em-4-carboxylate (125mg, 0.34mmol) in DMF (5ml) and pyridine (30 $\mu$ l, 0.37mmol). After work-up, the product was purified by triturating with diethyl ether to yield the title compound (148mg, 78%);  $\nu_{\text{max}}$  (KBr) 3343, 1751, 1718, 1678 and 1515 $\text{cm}^{-1}$ ;  $\delta_{\text{H}}$  ( $\text{CDCl}_3$ , 250MHz) 1.25-1.30 (2H, m), 1.50-2.78 (8H, m, 2H exch.), 3.75-3.95 (3H, m), 3.78 (3H, s), 4.12 (3H, s), 5.12 (1H, dd, J 7.8, 7.4Hz), 5.19 (2H, s), 5.43 (1H, dd, J 7.3, 5.1Hz), 6.88 (2H, d, J 8.7Hz), 7.05 (1H, s), 7.35 (2H, d, J 8.7Hz) and 8.09 (1H, br s, exch.). [Mass spectrum: +ve ion (3-nitrobenzyl alcohol, sodium acetate)  $\text{MH}^+$  (556),  $\text{MNa}^+$  (578)].

(g) Sodium (6RS,7SR)-7-[2-(aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(SR)-tetrahydrofuran-2-yl]-1-carba-1-dethiaceph-3-em-4-carboxylate

A solution of 4-methoxybenzyl (6RS,7SR)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(SR)-tetrahydrofuran-2-yl]-1-carba-1-dethiaceph-3-em-4-35 carboxylate (160mg, 0.29mmol) in dichloromethane (10ml) was

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added to a solution of aluminium chloride (115mg, 0.85mmol) in anisole (4.5ml) and dichloromethane (2.5ml) as described in Example 7(b). After quenching with trisodium citrate (0.5M, 9ml) and subsequent work-up, the product was purified by chromatography on HP20SS eluting with water, then 1 and 2% THF in water. Fractions containing the product (h.p.l.c. analysis) were combined and freeze-dried to give the title compound (94mg, 71%);  $\nu_{\text{max}}$  (KBr) 1745, 1663, 1595, 1532 and  $1387\text{cm}^{-1}$ ;  $\delta_{\text{H}}$  ( $d_6$ -DMSO, 250MHz) 1.38-1.55 (2H, m), 1.70-1.88 (3H, m), 1.97-2.16 (3H, m), 3.52-3.79 (3H, m), 3.82 (3H, s,), 4.95 (1H, dd,  $J$  8.4, 7.0Hz), 5.22 (1H, dd,  $J$  8.6, 4.9Hz), 6.73 (1H, s), 7.23 (2H, br s, exch.) and 9.18 (1H, d,  $J$  8.6Hz, exch.). [Mass spectrum: +ve ion (thioglycerol)  $\text{MH}^+$  (458)].

15 (h) Sodium (6RS,7SR)-7-[2-(2-aminothiazol-4-yl)-2-(2Z)-methoxyiminoacetamido]-3-[(RS)-tetrahydrofuran-2-yl]-1-carba-1-de thiaceph-3-em-4-carboxylate

A solution of 4-methoxybenzyl (6RS,7SR)-7-[2-(2-  
 20 aminothiazol-4-yl)-2-(Z)-methoxyaminoacetamido]-3-[(RS)-  
 tetrahydrofuran-2-yl]-1-carba-1-dethiaceph-3-em-4-  
 carboxylate (140mg, 0.25mmol) in dichloromethane (10ml) was  
 added to a solution of aluminium chloride (101mg, 0.76mmol)  
 in anisole (4.5ml) and dichloromethane (2.5ml) as described  
 25 in Example 7(b). After quenching with trisodium citrate  
 (0.5M, 8ml) and subsequent work-up, the product was purified  
 by chromatography on HP20SS eluting with water, then 1 and  
 2% THF in water. Fractions containing the product (h.p.l.c.  
 analysis) were combined and freeze-dried to give the title  
 30 compound (54mg, 47%);  $\nu_{\text{max}}$  (KBr) 1746, 1662, 1596, 1532 and  
 1387cm<sup>-1</sup>;  $\delta_{\text{H}}$  (d<sub>6</sub>-DMSO, 250MHz) 1.42-1.62 (2H,m), 1.68-1.88  
 (4H,m), 2.01 (1H,m), 2.27 (1H,m), 3.56-3.78 (3H,m), 3.85  
 (3H,s), 5.20 (2H,m), 6.75 (1H,s), 7.24 (2H,br s, exch.) and  
 9.25 (1H,d, J 8.7Hz). [Mass spectrum: +ve ion (thioglycerol)  
 35  $\text{MH}^+$  (458)].

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Example 25

Sodium (6R,7R)-7-[2-(2-Aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(S)-tetrahydrofuran-2-yl]ceph-3-em-4-5 carboxylate-1,1-dioxide

(a) 4-Methoxybenzyl (6R,7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(S)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate-1,1-dioxide

10 To an ice-cooled solution of 4-methoxybenzyl (6R,7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(S)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate (see Example 7) (300mg, 0.52mmol) in ethyl acetate (40ml) was added a 15 solution of m-chloroperbenzoic acid (270mg, 1.56mmol) in ethyl acetate (10ml). The solution was stirred at room temperature for 1h and was then washed with dilute aqueous sodium hydrogen carbonate and water, dried (magnesium sulphate) and evaporated under reduced pressure. The 20 residue was chromatographed on silica gel eluting with ethyl acetate/hexane mixtures to give the title compound as a cream coloured solid (50mg, 15%);  $\nu_{\text{max}}$  (CHCl<sub>3</sub>) 1810, 1730 and 1690cm<sup>-1</sup>;  $\delta_{\text{H}}$  (CDCl<sub>3</sub>) 1.52-1.70 (1H, m), 1.94-2.00 (2H, m), 2.41-2.48 (1H, m), 3.55 and 3.85 (2H, ABq,  $J$  19Hz), 3.19 25 (3H, s), 3.3-3.43 (2H, m), 4.1 (3H, s), 4.90 (1H, d,  $J$  5Hz), 4.97 (1H, t,  $J$  7Hz), 5.20 (2H, s), 5.94-6.3 (2H, m, exch.), 6.20 (1H, dd,  $J$  5, 10Hz), 6.91 (2H, d,  $J$  8Hz), 7.06 (1H, s), 7.32 (2H, d,  $J$  8Hz) and 7.86 (1H, d,  $J$  10Hz, exch). [Mass spectrum: +ve ion (thioglycerol)  $\text{MH}^+$  (606)].

30 (b) Sodium (6R,7R)-7-[2-(2-Aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(S)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate-1,1-dioxide

35 The product from Example 25(a) was treated by the method of Example 23(b) to give the title compound (51%) as a freeze-

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dried white solid;  $\nu_{\text{max}}$  (KBr) 1783, 1675 and 1610cm<sup>-1</sup>;  $\delta_{\text{H}}$  ( $d_6$ -DMSO) 1.45-1.50 (1H, m), 1.69-1.79 (2H, m), 2.00-2.11 (1H, m), 3.48 and 3.87 (2H, ABq,  $J$  18Hz), 3.76 (3H, s), 3.50-3.86 (2H, m), 4.85 (1H, t,  $J$  7Hz), 5.22 (H, d,  $J$  5Hz), 5.61 (1H, dd,  $J$  5, 7Hz), 6.79 (1H, s), 7.13 (2H, s, exch.) and 9.33 (1H, d,  $J$  7Hz exch.). [Mass spectrum: +ve ion (thioglycerol)  $\text{MNa}^+$  (508)].

Example 26

10

(RS)-1-(Propan-2-yl)oxycarbonyloxyethyl (6R,7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(S)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate

15 A solution of (RS)-1-iodo-1-(propan-2-yl)oxycarbonyl-oxoethane (516mg) in 1-methyl-2-pyrrolidinone (2ml) was added dropwise over 45mins to an ice-cold mixture of sodium (6R,7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(S)-tetrahydrofuran-2-yl]ceph-3-em-4-  
20 carboxylate (237mg) and finely powdered potassium carbonate (276mg) in 1-methyl-2-pyrrolidinone (5ml). The mixture was stirred for an additional 15mins, diluted with ethyl acetate, washed with water, brine, dried (magnesium sulphate), concentrated and flash chromatographed on silica  
25 gel eluting with 50, 60, 70, 80% ethyl acetate in hexane to give the title compound as a foam (58mg);  $\nu_{\text{max}}$  (CHCl<sub>3</sub>) 2960, 1787, 1760, 1682, 1633, 1519 and 1377cm<sup>-1</sup>;  $\delta$  (CDCl<sub>3</sub>, 250MHz) 1.20-2.50 (13H, m), 3.35-3.80 (2H, m), 3.80-4.20 (2H, m), 4.22 (3H, s), 4.83-5.10 (2H, m), 5.85-6.00 (1H, m), 6.85-  
30 7.08 (1H, m), 7.27 (1H, s) and 7.76 (1H, br, m). [Mass spectrum: +ve ion (3-nitrobenzyl alcohol, sodium acetate)  $\text{MNa}^+$  (606)].

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Example 27

Sodium (6R,7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxy-iminoacetamido]-3-[(5R,2SR)-5-methyltetrahydrofuran-2-5 yl]ceph-3-em-4-carboxylate

(a) Methyl 5-methyl-2-furoate

A solution of methyl 5-chloromethyl-2-furoate (5.0g,  
10 28.7mmol) in ethyl acetate (40ml) was hydrogenated over 10%  
palladium on charcoal (50mg) for 3h. The catalyst was  
filtered off and washed with ethyl acetate. The combined  
filtrates were concentrated in vacuo and the residue  
purified by chromatography on silica gel eluting with 10%  
15 ethyl acetate in hexane to yield the title compound as a  
colourless oil (3.78g, 94%);  $\nu_{\text{max}}$  ( $\text{CH}_2\text{Cl}_2$ ) 1725, 1534, 1522,  
1437 and  $1311\text{cm}^{-1}$ ;  $\delta_{\text{H}}$  ( $\text{CDCl}_3$ , 90MHz) 2.38 (3H, s), 3.86 (3H,  
s), 6.12 (1H, br d,  $J$  4Hz) and 7.07 (1H, d,  $J$  4Hz). [Mass  
spectrum:  $\underline{M}^+$  (140)].

20

(b) 5-Methyl-2-furoic acid

Methyl 5-methyl-2-furoate (3.68g, 26.29mmol) in methanol  
(30ml) was treated with a solution of potassium hydroxide  
25 (2.80g, 50.0mmol) in water (15ml) and the mixture stirred  
for 2h at room temperature. The methanol was evaporated in  
vacuo, the residue dissolved in water and washed with ethyl  
acetate. The aqueous phase was acidified with 5N  
hydrochloric acid, and the product extracted with ethyl  
30 acetate (x3). The combined organic solutions were dried and  
concentrated to yield the title compound as a yellow solid  
(3.12g, 94%); m.p. 110-112°C; (Found:  $\underline{M}^+$ , 126.0312.  $\text{C}_6\text{H}_6\text{O}_3$   
requires  $\underline{M}^+$  126.0317);  $\nu_{\text{max}}$  ( $\text{CH}_2\text{Cl}_2$ ) 3300-2700, 1688, 1524,  
1424, 1305, 1210 and  $1167\text{cm}^{-1}$ ;  $\delta_{\text{H}}$  ( $\text{CDCl}_3$ , 90MHz) 2.40 (3H,  
35 s), 6.15 (1H, d,  $J$  4Hz) and 7.22 (1H, d,  $J$  4Hz).

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(c) 5-Methyl-2-tetrahydrofuroic acid

A solution of 5-methyl-2-furoic acid (3.65g, 28.97mmol) in ethyl acetate (60ml) was hydrogenated over 5% rhodium on 5 carbon (250mg) until hydrogen uptake ceased. The catalyst was filtered off and washed with ethyl acetate. The combined filtrates were concentrated in vacuo to yield the title compound as a pale yellow oil (3.67g, 97%);  $\nu_{\text{max}}$  ( $\text{CH}_2\text{Cl}_2$ ) 3384, 3359, 1775, 1724 and  $1355\text{cm}^{-1}$ ;  $\delta_{\text{H}}$  ( $\text{CDCl}_3$ , 10 250MHz) 1.35 (3H, d,  $J$  6.1Hz), 1.53 (1H, m), 2.09 (1H, m), 2.17-2.40 (2H, m), 4.21 (1H, m) and 4.46 (1H, dd,  $J$  8.9, 4.7Hz). [Mass spectrum: +ve ion (ammonia)  $\text{MNH}_4^+$  (148)].

(d) 2-Bromoacetyl-5-methyltetrahydrofuran

15 A solution of 5-methyl-2-tetrahydrofuroic acid (1.80g, 13.85mmol) in dichloromethane (25ml) was treated with oxalyl chloride (2.4ml, 27.51mmol) in the presence of dimethylformamide (3 drops). After stirring for 1.25h, the 20 solvent was evaporated in vacuo. The residue was re-dissolved in dichloromethane and concentrated again. Excess diazomethane was then bubbled through a solution of the resulting acid chloride in dichloromethane (30ml) at  $0^\circ\text{C}$ . When the addition was complete, the mixture was stirred for 25 10min. at  $0^\circ\text{C}$  and then treated with 48% aqueous hydrogen bromide (2.6ml, 15.41mmol). The mixture was stirred for 15min. at room temperature, washed with water (x2), dried and concentrated in vacuo to yield the crude title compound as a brown oil (1.67g, 58%);  $\nu_{\text{max}}$  ( $\text{CH}_2\text{Cl}_2$ ) 1735, 1387 and 30  $1086\text{cm}^{-1}$ ;  $\delta_{\text{H}}$  ( $\text{CDCl}_3$ , 90MHz) 1.33 (3H, d,  $J$  6.0Hz), 1.48 (1H, m), 1.90-2.35 (3H, m), 4.10 (1H, m), 4.25 (2H, s) and 4.48 (1H, m).

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(e) 4-Methoxybenzyl (2RS)-2-hydroxy-2-[(3R,4R)-4-(5-methyltetrahydrofuran-2-ylcarbonylmethylthio)-3-phenylacetamidoazetidin-2-on-1-yl]acetate

5 Toluene-4-sulphonic acid (3.42g, 17.98mmol) in water (8ml) was added to a solution of 4-methoxybenzyl (2RS)-2-hydroxy-2-[(1R,5R)-3-benzyl-4-thia-2,6-diazabicyclo[3.2.0]hept-2-en-7-on-6-yl]acetate (4.12g, 10.0mmol) in dichloromethane (20ml) and acetone (20ml). After stirring for 2.5h at room 10 temperature, the reaction mixture was diluted with dichloromethane, washed with water (x2), dried and concentrated in vacuo to yield crude 4-methoxybenzyl (2RS)-2-hydroxy-2-[(3R,4R)-4-mercaptop-3-phenylacetamidoazetidin-2-on-1-yl]acetate as a colourless foam. The crude thiol was 15 dissolved in acetone (50ml) and treated with a solution of 2-bromoacetyl-5-methyltetrahydrofuran (1.67g, 8.1mmol) in acetone (5ml). After 10min., potassium carbonate (687mg, 5.0mmol) was added, and the mixture stirred for a further 30min. The reaction mixture was diluted with ethyl acetate, 20 washed successively with water (x2) and brine, dried and concentrated. The residue was purified by chromatography on silica gel eluting with 50, 70 and 80% ethyl acetate in hexane to yield the title compound as a colourless foam (2.68g, 60%);  $\nu_{\text{max}}$  ( $\text{CH}_2\text{Cl}_2$ ) 3412, 1781, 1744, 1685 and 25  $1515\text{cm}^{-1}$ . [Mass spectrum: +ve ion (3-nitrobenzyl alcohol, sodium acetate)  $\text{MNa}^+$  (579)].

(f) 4-Methoxybenzyl 2-[(3R,4R)-4-(5-methyltetrahydrofuran-2-ylcarbonylmethylthio)-3-phenylacetamidoazetidin-2-on-1-yl]-2-tri-n-butylphosphoranylideneacetate

A solution of thionyl chloride (530 $\mu$ l, 7.27mmol) in tetrahydrofuran (5ml) was added dropwise to the hydroxy compound (2.68g, 4.85mmol) and 2,6-lutidine (850 $\mu$ l, 35 7.29mmol) in tetrahydrofuran (30ml) at -20°C. After stirring for 30min. the reaction mixture was filtered through a pad of celite and the filtrate concentrated in

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vacuo. Toluene was added and re-evaporated to yield 4-methoxybenzyl (RS)-2-chloro-2-[(3R, 4R)-4-(5-methyltetrahydrofuran-2-ylcarbonylmethylthio)-3-phenyl-acetamido-azetidin-2-on-1-yl]acetate. The crude chloro-compound was dissolved in dioxan (40ml) and treated with tri-n-butylphosphine (2.7ml, 10.84mmol). After stirring for 30min. at room temperature, the reaction mixture was diluted with ethyl acetate and washed successively with dilute sodium hydrogen carbonate solution, water and brine. The organic solution was dried, concentrated and then purified by chromatography on silica gel eluting with 50, 70 and 100% ethyl acetate in hexane to yield the title compound as a yellow foam (2.28g, 64%);  $\nu_{\text{max}}$  ( $\text{CH}_2\text{Cl}_2$ ) 3420, 1762, 1732, 1681 and  $1515\text{cm}^{-1}$ . [Mass spectrum: +ve ion (3-nitrobenzyl alcohol, sodium acetate)  $\text{MH}^+$  741,  $\text{MNa}^+$  763].

(g) 4-Methoxybenzyl (6R, 7R)-3-(5-methyltetrahydrofuran-2-yl)-7-phenylacetamidoceph-3-em-4-carboxylate

20 A solution of the phosphorane (2.28g, 3.08mmol) and benzoic acid (10mg) in toluene (40ml) was heated in an oil bath at 130°C for 16h under argon. The reaction mixture was cooled, concentrated and the residue purified by chromatography on silica gel eluting with 10, 20 and 40% ethyl acetate in hexane yielding a mixture of the title compound and some of the Δ2 isomer as a yellow oil (1.27g, 79%); (Found:  $M^+$ , 522.1813.  $C_{28}H_{30}N_2O_6S_2$  requires  $M^+$  522.1825);  $\nu_{max}$  ( $CH_2Cl_2$ ) 3416, 1782, 1729, 1688, 1613 and  $1515\text{cm}^{-1}$ .

30 (h) 4-Methoxybenzyl (6R,7R)-7-amino-3-(5-methyl-tetrahydrofuran-2-yl)ceph-3-em-4-carboxylate

Phosphorus pentachloride (754mg, 3.62mmol) in dichloromethane (19ml) was added to 4-methoxybenzyl (6*R*, 7*R*)-35 3-(5-methyltetrahydrofuran-2-yl)-7-phenylacetamidoceph-3-em-

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4-carboxylate (containing some of the  $\Delta 2$ -isomer) (1.26g, 2.41mmol) and N-methylmorpholine (531 $\mu$ l, 4.83mmol) in dichloromethane (15ml) at -25°C. The reaction was stirred at -10 $\pm$ 5°C for 45min., then methanol (5ml) was added, and stirring was continued for 45min. at room temperature. Water (10ml) was then added, and the mixture vigorously stirred for a further 1h. After evaporation of the dichloromethane in vacuo, the pH of the aqueous residue was adjusted to 7 by the addition of ammonium hydroxide in the presence of ethyl acetate. The mixture was extracted with ethyl acetate (x2), dried and concentrated in vacuo. The residue was purified by chromatography on silica gel eluting with 30, 50, 70, 80 and 100% ethyl acetate in hexane yielding 4-methoxybenzyl (6R,7R)-7-amino-3-[(5S,2S)-5-methyltetrahydrofuran-2-yl]ceph-3-em-4-carboxylate (310mg, 32%) as a pale yellow foam; (Found:  $M^+$ , 404.1394.  $C_{20}H_{24}N_2O_5S$  requires  $M^+$  404.1406);  $\nu_{max}$  ( $CH_2Cl_2$ ) 3412, 1776, 1721, 1613, 1516 and 1393 $cm^{-1}$ ;  $\delta_H$  ( $CDCl_3$ , 250MHz), 1.24 (3H, d,  $\underline{\Delta}$  5.8Hz), 1.48 (1H, m), 1.69 (1H, m), 2.02 (3H, m, 2H exch.), 2.25 (1H, m), 3.45 and 3.60 (2H, ABq,  $\underline{\Delta}$  17.7Hz), 3.78 (3H, s), 3.98 (1H, m), 4.88 (1H, d,  $\underline{\Delta}$  5.0Hz), 4.93-5.04 (2H, m), 5.17 (2H, s), 6.87 (2H, d,  $\underline{\Delta}$  8.6Hz), 7.32 (2H, d,  $\underline{\Delta}$  8.6Hz).

Further elution of the column with ethyl acetate yielded the more polar diastereoisomer 4-methoxybenzyl (6R,7R)-7-amino-3-[(5R,2R)-5-methyltetrahydrofuran-2-yl]ceph-3-em-4-carboxylate (208mg, 21%) as a yellow foam; (Found:  $M^+$  404.1402.  $C_{20}H_{24}N_2O_5S$  requires  $M^+$  404.1406);  $\nu_{max}$  ( $CH_2Cl_2$ ) 3411, 1776, 1727, 1613 and 1516 $cm^{-1}$ ;  $\delta_H$  ( $CDCl_3$ , 250MHz), 1.24 (3H, d,  $\underline{\Delta}$  6.1Hz), 1.48 (1H, m), 1.69 (1H, m), 1.92-2.08 (2H, m), 3.47 and 3.71 (2H, ABq,  $\underline{\Delta}$  17.8Hz), 3.79 (3H, s), 4.00 (1H, dd,  $\underline{\Delta}$  12.9, 6.4Hz), 4.83 (1H, d,  $\underline{\Delta}$  4.8Hz), 4.92-5.17 (4H, m, 2H exch.), 5.19 (2H, s), 6.88 (2H, d,  $\underline{\Delta}$  8.6Hz) and 7.32 (2H, d,  $\underline{\Delta}$  8.6Hz).

Further elution of the column yielded the  $\Delta 2$ -cephems (142mg, 15%).

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(i) 4-Methoxybenzyl (6R,7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(5S,2S)-5-methyltetrahydrofuran-2-yl]ceph-3-em-4-carboxylate

5 2-(2-Aminothiazol-4-yl)-2-(Z)-methoxyiminoacetic acid (167mg, 0.83mmol) in DMF (5ml) was treated with methanesulphonyl chloride (64 $\mu$ l, 0.83mmol) and N,N-diisopropylethylamine (145 $\mu$ l, 0.83mmol) as described in Example 7(a). This was then treated successively with a 10 solution of 4-methoxybenzyl (6R,7R)-7-amino-3-[(5S,2S)-5-methyltetrahydrofuran-2-yl]ceph-3-em-4-carboxylate (305mg, 0.75mmol) in DMF (5ml) and pyridine (67 $\mu$ l, 0.83mmol). After work-up the product was purified by chromatography on silica gel eluting with 50, 70 and 100% ethyl acetate in hexane to 15 yield the title compound as a yellow foam (373mg, 85%);  $\nu_{\text{max}}$  ( $\text{CH}_2\text{Cl}_2$ ) 3389, 1784, 1724, 1689, 1606 and 1516 $\text{cm}^{-1}$ ;  $\delta_{\text{H}}$  ( $\text{CDCl}_3$ , 400MHz) 1.29 (3H, d,  $\underline{\text{J}}$  5.9Hz), 1.48 (1H, m), 1.69 (1H, m), 1.93 (2H, br s, exch.), 2.07 (1H, m), 2.29 (1H, m), 3.39 and 3.64 (2H, ABq,  $\underline{\text{J}}$  18.8Hz), 3.80 (3H, s), 4.00 (1H, 20 dd,  $\underline{\text{J}}$  12.8, 6.4Hz), 4.10 (3H, s), 4.96 (1H, dd,  $\underline{\text{J}}$  7.7, 7.7Hz), 5.02 (1H, d,  $\underline{\text{J}}$  4.8Hz), 5.19 (2H, s), 5.84 (1H, br s, exch.), 5.94 (1H, dd,  $\underline{\text{J}}$  9.0, 4.8Hz), 6.89 (2H, d,  $\underline{\text{J}}$  8.5Hz) and 7.02 (1H, s), 7.35 (2H, d,  $\underline{\text{J}}$  8.5Hz). [Mass spectrum: +ve ion (ammonia)  $\text{MH}^+$  (588)].

25

(j) 4-Methoxybenzyl (6R,7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(5R,2R)-5-methyltetrahydro-furan-2-yl]ceph-3-em-4-carboxylate

30 2-(2-Aminothiazol-4-yl)-2-(Z)-methoxyiminoacetic acid (109mg, 0.54mmol) in DMF (3ml) was treated with methanesulphonyl chloride (42 $\mu$ l, 0.54mmol) and N,N-diisopropylethylamine (95 $\mu$ l, 0.55mmol) as described in Example 7(a). This was then treated successively with a 35 solution of 4-methoxybenzyl (6 $R$ , 7 $R$ )-7-amino-3-[(5 $R$ , 2 $R$ )-5-methyltetrahydrofuran-2-yl]ceph-3-em-4-carboxylate (200mg, 0.50mmol) in DMF (10ml) and pyridine (44 $\mu$ l, 0.54mmol).

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After work-up, the product was purified by triturating with diethyl ether to yield the title compound (214mg, 73%);  $\nu_{\text{max}}$  (CH<sub>2</sub>Cl<sub>2</sub>) 3388, 1784, 1726, 1688, 1606 and 1516cm<sup>-1</sup>;  $\delta_{\text{H}}$  (CDCl<sub>3</sub>, 400MHz) 1.26 (3H, d,  $J$  6.0Hz), 1.46 (1H, m), 1.66 (1H, m), 1.87 (2H, br s, exch.), 2.00 (2H, m), 3.43 and 3.67 (2H, ABq,  $J$  18.0Hz), 3.81 (3H, s), 4.00 (1H, dd,  $J$  13.3, 6.3Hz), 4.09 (3H, s), 5.04 (1H, d,  $J$  4.8Hz), 5.15-5.25 (3H, m), 5.55 (1H, br s, exch.), 5.89 (1H, dd,  $J$  8.8, 4.8Hz), 6.90 (2H, d,  $J$  8.6Hz), 6.98 (1H, s) and 7.34 (2H, d,  $J$  8.6Hz). [Mass spectrum: +ve ion (ammonia)  $\text{MH}^+$  (588)].

(k) Sodium (6R,7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(5S,2S)-5-methyltetrahydrofuran-2-yl]ceph-3-em-4-carboxylate

15 A solution of 4-methoxybenzyl (6R,7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(5S,2S)-5-methyltetrahydrofuran-2-yl]ceph-3-em-4-carboxylate (370mg, 0.63mmol) in dichloromethane (10ml) was added to a solution 20 of aluminium chloride (252mg, 1.89mmol) in anisole (10ml) and dichloromethane (5ml) as described in Example 7(b). After quenching with trisodium citrate (0.5M, 20ml) and subsequent work-up, the product was purified by chromatography on HP20SS eluting with water, then 1, 2, 3 25 and 4% THF in water. Fractions containing the product (h.p.l.c. analysis) were combined and freeze-dried to give the title compound (240mg, 78%);  $\nu_{\text{max}}$  (KBr) 1762, 1670, 1602, 1532 and 1390cm<sup>-1</sup>;  $\delta_{\text{H}}$  (d<sub>6</sub>-DMSO, 250MHz) 1.15 (3H, d,  $J$  6.0Hz), 1.41 (1H, m), 1.59 (1H, m), 1.85-2.08 (2H, m), 3.26 (2H, ABq,  $J$  17.7Hz), 3.85 (3H, s), 3.87 (1H, m), 4.87 (1H, dd,  $J$  7.3, 7.3Hz), 5.00 (1H, d,  $J$  4.7Hz), 5.57 (1H, dd,  $J$  8.0, 4.7Hz), 6.74 (1H, s), 7.22 (2H, s, exch.) and 9.51 (1H, d,  $J$  8.0Hz, exch.). [Mass spectrum: +ve ion (thioglycerol)  $\text{MH}^+$  (490)].

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(1) Sodium (6R, 7R)-7-[2-(2-aminothiazol)-4-yl)-2-(2-methoxyiminoacetamido]-3-[(5R, 2SR)-5-methyltetrahydrofuran-2-yl]ceph-3-em-4-carboxylate

5 4-Methoxybenzyl (6*R*,7*R*)-7-[2-(2-aminothiazol-4-yl)-2-(*Z*)-methoxyiminoacetamido]-3-[*(5R,2R)*-5-methyltetrahydrofuran-2-yl]ceph-3-em-4-carboxylate (210mg, 0.36mmol) in 0.1M hydrochloric acid in 90% formic acid (3.6ml) was allowed to stand for 1h. Concentrated hydrochloric acid (2 drops) was 10 then added, and the mixture left for a further 2.5h. After evaporating to dryness in vacuo, the residue was dissolved in water, the pH adjusted to 6.5 by addition of 1M sodium hydroxide solution and chromatographed on HP20SS eluting with 0, 1, 2, 3 and 4% THF in water. Fractions containing 15 the product (h.p.l.c. analysis) were combined, concentrated and freeze-dried to give the title compound as a mixture of diastereoisomers (121mg, 69%);  $\nu_{\text{max}}$  (KBr) 1763, 1663, 1598 and  $1388\text{cm}^{-1}$ ;  $\delta_{\text{H}}$  ( $d_6$ -DMSO, 250MHz) 1.10 and 1.16 (together 3H, 2d,  $\underline{\text{J}}$  6.0Hz), 1.27-2.17 (4H, m), 3.15-3.45 (together 2H, 20 2ABq), 3.84 (3H, s), 4.09 (1H, m), 4.93 and 4.95 (together 1H, 2d,  $\underline{\text{J}}$  4.6Hz), 5.02 and 5.18 (together 1H, 2dd,  $\underline{\text{J}}$  9.4, 5.9 and 7.6, 7.6Hz), 5.50 (1H, m), 6.74 and 6.76 (together 1H, 2s), 7.22 (2H, s, exch.) and 9.47 and 9.52 (together 1H, 2d,  $\underline{\text{J}}$  8.4Hz, exch.). [Mass spectrum: +ve ion (thioglycerol) 25  $\text{MH}^+$  (490)].

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EXAMPLE 28

Sodium (6R,7R)-7-[2-(furan-2-yl)-2-(Z)-methoxyimino-  
acetamido]-3-[(S)-tetrahydrofuran-2-yl]ceph-3-em-4-  
5 carboxylate

(a) 4-Methoxybenzyl (6R,7R)-7-[2-(furan-2-yl)-2-(Z)-  
methoxyiminoacetamido]-3-[(S)-tetrahydrofuran-2-yl]ceph-3-  
em-4-carboxylate

10 2-(Furan-2-yl)-2-(Z)-methoxyiminoacetic acid (90mg) in dry  
DMF (4ml) was treated with N,N-diisopropylethylamine  
(0.1ml), cooled to -35°C, and treated with methanesulphonyl  
chloride (0.044ml) and the mixture stirred at -35°C for  
15 30min.

20 A solution of the 4-methoxybenzyl (6R,7R)-7-amino-3-[(S)-  
tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate (195mg) in dry  
DMF (3ml) was added followed by pyridine (0.044ml) and the  
mixture stirred at ice-bath temperature for a further 1h.  
The solution was diluted with excess ethyl acetate and the  
organic solution washed successively with 5% aqueous citric  
acid, saturated aqueous sodium bicarbonate solution and  
finally brine. After drying over anhydrous magnesium  
25 sulphate the solvent was evaporated. Chromatography of the  
residue on silica gel using ethyl acetate-hexane (1:1) as  
eluent gave the title compound as a pale yellow foam (190mg,  
73%);  $\nu_{\text{max}}$  3400, 1785, 1725 and 1690cm<sup>-1</sup>. [Mass spectrum:  
+ve ion (thioglycerol)  $\text{MH}^+$  (542)].

30 (b) Sodium (6R,7R)-7-[2-furan-2-yl)-2-(Z)-methoxyimino-  
acetamido]-3-[(S)-tetrahydrofuran-2-yl]ceph-3-em-4-  
carboxylate

35 Aluminium trichloride (130mg) was added to a solution of  
anisole (6ml) and dichloromethane (4ml) at -25°C and the  
mixture stirred at -25°C for 15min. The mixture was then  
cooled to -40°C, a solution of the product of Example 28(a)

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(180mg) in dichloromethane (4ml) added in one portion and stirred at -40° for 20min. The cooling bath was removed, trisodium citrate (10ml of an aqueous 0.5M solution) added and the mixture stirred vigorously for 20min. The aqueous layer was separated, washed twice with dichloromethane and concentrated under reduced pressure. The residue was chromatographed on HP20SS eluting with water-acetone mixtures. Fractions containing the product (t.l.c.; h.p.l.c. analysis) were combined, concentrated and freeze-dried to give the title compound as a white solid (95mg, 66%);  $\mu_{\text{max}}$  (KBr) 1770, 1685 and 1600cm<sup>-1</sup>;  $\delta_{\text{H}}$  (D<sub>2</sub>O) 1.65-1.85 (1H, m), 1.9-2.05 (2H, m), 2.08-2.15 (1H, m), 3.33 and 3.53 (2H, ABq,  $J$  18Hz), 3.75-4.0 (2H, m), 3.96 (3H, s), 4.71 (1H, dd,  $J$  8.3, 6.9Hz), 5.2 (1H, d,  $J$  4.5Hz), 5.73 (H, d,  $J$  4.5Hz), 6.58 (1H, dd), 6.86 (1H, d) and 7.64 (1H, d).

EXAMPLE 29

Sodium (6R,7R)-7-[2-(2-aminothiazol)-2-(Z)-methoxyimino-acetamido]-3-[(S)-5,5-dimethyltetrahydrofuran-2-yl]ceph-3-em-4-carboxylate

(a) (S)-2-Bromoacetyl-5,5-dimethyltetrahydrofuran

A solution of (S)-5,5-dimethyltetrahydrofuran-2-carboxylic acid (800mg, 5.56mmol) (I. Kitagawa, T. Nishino, M. Kobayashi, T. Matsuno, H. Akutsu and Y. Kyagaku, Chem. Pharm. Bull., 1981, 29, 1942) in dichloromethane (25ml) was treated with oxalyl chloride (2.4ml, 27.51mmol) and dimethylformamide (3 drops). The mixture was stirred for 1h, evaporated in vacuo, dichloromethane added, and re-evaporated. The resulting acid chloride was dissolved in dichloromethane (25ml) and cooled in an ice-bath. Diazomethane was then passed into the solution as described in Example 14(a). When the addition was complete, 48% aqueous hydrogen bromide (2.6ml) was added, and the mixture

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stirred for a further 10min. The solution was washed with water (x2), dried over  $MgSO_4$  and concentrated in vacuo to yield the title compound as an orange oil (812mg, 66%);  $\nu_{max}$  ( $CH_2Cl_2$ )  $1767cm^{-1}$ ;  $\delta_H$  ( $CDCl_3$ , 250MHz) 1.28 (3H, s), 1.32 (3H, s), 1.78-2.68 (4H, m), 4.27 (2H, s) and 4.56 (1H, dd,  $J$  8.2, 6.8Hz).

(b) 4-Methoxybenzyl (2RS)-2-Hydroxy-2-[(3R,4R)-4-[(S)-5,5-dimethyltetrahydrofuran-2-ylcarbonylmethylthio]-3-phenylacetamidoazetidin-2-on-1-yl]acetate

4-Methoxybenzyl (RS)-2-hydroxy-2-[(1R,5R)-3-benzyl-4-thia-2,6-diazabicyclo[3.2.0]hept-2-en-7-on-6-yl]acetate (3.3g, 8.0mmol) in 50% acetone/dichloromethane (32ml) was cleaved 15 with 4-toluenesulphonic acid (2.74g, 14.4mmol) in water (6ml). This product was reacted with the crude bromide from Example 29(a) (808mg, 3.66mmol) in acetone (40ml) with potassium carbonate (550mg, 3.99mmol) as described in Example 6(b). After work-up, the residue was purified by 20 chromatography on silica gel eluting with 50, 70 and 90% ethyl acetate in hexane to yield the title compound (1.25g, 60%) as a yellow oil;  $\nu_{max}$  ( $CH_2Cl_2$ ) 3410, 1780, 1746, 1683, 1613 and  $1515cm^{-1}$ . [Mass spectrum: +ve ion (3-nitrobenzyl alcohol, sodium acetate)  $MNa^+$  (593)].

25

(c) 4-Methoxybenzyl 2-[(3R,4R)-4-[(S)-5,5-dimethyltetrahydrofuran-2-ylcarbonylmethylthio]-3-phenylacetamidoazetidin-2-on-1-yl]-2-tri-n-butylphosphoranylideneacetate

30

The alcohol from Example 29(b) (1.25g, 2.19mmol) was treated with thionyl chloride (240 $\mu$ l, 3.29mmol) and 2,6-lutidine (383 $\mu$ l, 3.29mmol), followed by tri-n-butylphosphine (1.20ml, 4.82mmol) as described for Example 6(c). The product was 35 purified by chromatography on silica gel eluting with 50, 70

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and 100% ethyl acetate in hexane to yield the title compound (617mg, 37%) as a yellow foam;  $\nu_{\text{max}}$  (CH<sub>2</sub>Cl<sub>2</sub>) 1763, 1680, 1608 and 1515cm<sup>-1</sup>. [Mass spectrum:  $\underline{M}^+$  (754)].

5 (d) 4-Methoxybenzyl (6R,7R)-3-[(S)-5,5-dimethyltetrahydrofuran-2-yl]-7-phenylacetamidoceph-3-em-4-carboxylate

A solution of the phosphorane from Example 29(c) (610mg, 0.81mmol) and benzoic acid (10mg) in toluene (20ml) was 10 heated at reflux for 16h. After cooling, the solvent was evaporated in vacuo. The residue was purified by chromatography on silica gel eluting with 5 and 10% ethyl acetate in dichloromethane yielding the title compound as a yellow foam (240mg, 55%); (Found:  $\underline{M}^+$ , 536.1978. C<sub>29</sub>H<sub>32</sub>N<sub>2</sub>O<sub>6</sub>S 15 requires  $\underline{M}^+$  536.1981);  $\nu_{\text{max}}$  (CH<sub>2</sub>Cl<sub>2</sub>) 3415, 1784, 1723, 1684 and 1515cm<sup>-1</sup>;  $\delta_{\text{H}}$  (CDCl<sub>3</sub>, 250MHz) 1.22 (3H, s), 1.27 (3H, s), 1.62-1.81 (3H, m), 2.28 (1H, m), 3.30 and 3.56 (2H, ABq,  $\underline{J}$  18.8Hz), 3.60 and 3.69 (2H, ABq,  $\underline{J}$  16.3Hz), 3.82 (3H, s), 4.88 (1H, d,  $\underline{J}$  4.8Hz), 5.00 (1H, dd,  $\underline{J}$  8.6, 6.1Hz), 5.11 and 20 5.21 (2H, ABq,  $\underline{J}$  11.8Hz), 5.80 (1H, dd,  $\underline{J}$  9.1, 4.8Hz), 5.96 (1H, br d,  $\underline{J}$  9.1Hz, exch.), 6.88 (2H, d,  $\underline{J}$  8.7Hz) and 7.24- 7.40 (7H, m).

(e) 4-Methoxybenzyl (6R,7R)-7-amino-3-[(S)-5,5-dimethyltetrahydrofuran-2-yl]ceph-3-em-4-carboxylate

Phosphorus pentachloride (48mg, 0.23mmol) in dichloromethane (1.2ml) was added to 4-methoxybenzyl (6R,7R)-3-[(S)-5,5-dimethyltetrahydrofuran-2-yl]-7-phenylacetamidoceph-3-em-4- 30 carboxylate (93mg, 0.15mmol) and N-methylmorpholine (34 $\mu$ l, 0.31mmol) in dichloromethane (3ml) at -25°C. The reaction was stirred at -10±5°C for 45min., then methanol (0.5ml) was added, and stirring continued for 45min. at room temperature. Water (1ml) was then added, and the mixture 35 vigorously stirred for a further 1h. After evaporation of the dichloromethane in vacuo, the pH of the aqueous residue

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was adjusted to 7 by the addition of ammonium hydroxide in the presence of ethyl acetate. The mixture was extracted with ethyl acetate (x2), dried and concentrated in vacuo. The residue was purified by chromatography on silica gel eluting with 70% ethyl acetate in hexane yielding the title compound (25mg, 39%); (Found:  $M^+$  418.1566.  $C_{21}H_{26}N_2O_5S$  requires  $M^+$  418.1562);  $\nu_{max}$  ( $CH_2Cl_2$ ) 2970, 1777, 1721, 1613 and  $1516cm^{-1}$ ;  $\delta_H$  ( $CDCl_3$ , 250MHz) 1.21 (3H, s), 1.27 (3H, s), 1.68-1.81 (3H, m), 2.25 (1H, m), 3.48 and 3.62 (2H, ABq,  $J$  18.7Hz), 3.56 (2H, br s, exch.), 3.79 (3H, s), 4.73-5.25 (5H, m), 6.87 (2H, d,  $J$  8.6Hz) and 7.30 (2H, d,  $J$  8.6Hz).

(f) 4-Methoxybenzyl (6R,7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(S)-5,5-dimethyltetrahydrofuran-2-yl]ceph-3-em-4-carboxylate

2-(2-Aminothiazol-4-yl)-2-(Z)-methoxyiminoacetic acid (13mg, 0.065mmol) in DMF (2ml) was treated with methanesulphonyl chloride (5 $\mu$ l, 0.064mmol) and N,N-diisopropylethylamine (11 $\mu$ l, 0.063mmol) as described in Example 7(a). This was then treated successively with a solution of the amine from Example 29(e) (25mg, 0.060mmol) in DMF (2ml) and pyridine (5 $\mu$ l, 0.062mmol). After work-up the product was purified by chromatography on silica gel eluting with 50, 70 and 100% ethyl acetate in hexane to yield the title compound (25mg, 70%) as a yellow foam;  $\nu_{max}$  ( $CH_2Cl_2$ ) 3389, 1784, 1722, 1690, 1607 and  $1516cm^{-1}$ ;  $\delta_H$  ( $CDCl_3$ , 250MHz) 1.23 (3H, s), 1.29 (3H, s), 1.61-1.84 (3H, m), 2.31 (1H, m), 3.40 and 3.63 (2H, ABq,  $J$  18.7Hz), 3.81 (3H, s), 4.20 (3H, s), 4.99 (1H, d,  $J$  4.8Hz), 5.05 (1H, dd,  $J$  8.1, 8.1Hz), 5.13 and 5.23 (2H, ABq,  $J$  11.8Hz), 5.90 (1H, dd,  $J$  8.9, 4.8Hz), 6.90 (2H, d,  $J$  8.7Hz), 7.23 (1H, s), 7.34 (2H, d,  $J$  8.7Hz), 7.50 (2H, br s, exch.) and 7.68 (1H, br d,  $J$  8.9Hz, exch). [Mass spectrum: +ve ion (ammonia)  $MH^+$  (602)].

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(g) Sodium (6R, 7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(S)-5,5-dimethyltetrahydrofuran-2-yl]ceph-3-em-4-carboxylate

5 A solution of the ester from Example 29(f) (23mg, 0.038mmol) in dichloromethane (2ml) was added to a solution of aluminium chloride (15mg, 0.112mmol) in anisole (0.6ml) and dichloromethane (0.3ml) as described in Example 7(b). After quenching with trisodium citrate (0.5M, 1.3ml) and 10 subsequent work-up, the product was purified by chromatography on HP20SS eluting with water, then 1, 2, 4 and 6% THF in water. Fractions containing the product (h.p.l.c. analysis) were combined and freeze-dried to give the title compound (13mg, 68%);  $\nu_{\text{max}}$  (KBr) 1762, 1664, 1605 15 and  $1529\text{cm}^{-1}$ ;  $\delta_{\text{H}}$  ( $d_6$ -DMSO, 250MHz) 1.13 (3H, s), 1.19 (3H, s), 1.59-1.73 (3H, m), 2.04 (1H, m), 3.22 and 3.37 (2H, ABq,  $J$  17.5Hz), 3.83 (3H, s), 4.93 (1H, d,  $J$  4.5Hz), 5.00 (1H, dd,  $J$  7.9Hz), 5.52 (1H, dd,  $J$  8.0, 4.5Hz), 6.75 (1H, s), 7.23 (2H, br s, exch.) and 9.48 (1H, d,  $J$  8.0Hz, exch.). 20 [Mass spectrum: +ve ion (3-nitrobenzyl alcohol, sodium acetate)  $\text{MNa}^+$  (526)].

EXAMPLE 30

25 Sodium (6R, 7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-(5-methoxycarbonyltetrahydrofuran-2-yl]ceph-3-em-4-carboxylate

(a) (2RS, 5SR)-5-Methoxycarbonyltetrahydrofuran-2-yl carboxylic acid

A mixture of furan-2,5-dicarboxylic acid monomethyl ester (1.95g) and 5% rhodium on carbon (400mg) in ethyl acetate (50ml) was hydrogenated until hydrogen uptake ceased. The 35 catalyst was filtered off and washed with ethyl acetate. The combined filtrates were evaporated to give (2.00g) of

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the title compound:  $\nu_{\text{max}}$  (film) 3170, 1765 and 1720  $\text{cm}^{-1}$ ;  $\delta_{\text{H}}$  ( $\text{CDCl}_3$ ) 1.95-2.65 (4H, m), 3.85 (3H, s) and 4.55-4.8 (2H, m).

5 (b) Methyl (2RS,5SR)-5-(2-chloroacetyl)tetrahydro-2-furoate

Oxalyl chloride (1.55ml) was added to a stirred solution of (2RS,5SR)-5-methoxycarbonyltetrahydrofuran-2-ylcarboxylic acid (2.00g) in dichloromethane (30ml). Dimethylformamide (1 drop) was added and the mixture stirred at room temperature for 1h, and then heated to reflux for 10min. The mixture was cooled and the solvent removed on a rotary evaporator. Chloroform was then evaporated from the residue twice. The residue was dissolved in dichloromethane (100ml) and the solution cooled in an ice bath, then excess diazomethane was passed into the solution. The mixture was stirred at 0°C for 15min and then excess hydrogen chloride was passed into the solution. The solution was washed with brine, dried over magnesium sulphate and evaporated. The title compound (2.02g) was isolated by column chromatography of the residue using gradient elution (silica gel, 4:1 going to 1:1 hexane:ethyl acetate);  $\nu_{\text{max}}$  ( $\text{CHCl}_3$ ) 1740  $\text{cm}^{-1}$ ;  $\delta_{\text{H}}$  ( $\text{CDCl}_3$ ) 1.9-2.5 (4H, m), 3.71 (3H, s), 4.45-4.8 (2H, m), 25 4.54 (1H, d,  $\underline{\lambda}$  18Hz) and 4.89 (1H, d,  $\underline{\lambda}$  18Hz).

(c) (3R,4R)-4-[(2RS,5SR)-5-Methoxycarbonyltetrahydrofuran-2-ylcarbonylmethylthio)-3-phenylacetamidoazetidin-2-one

30 Potassium carbonate (2.0g) was added to a stirred solution of (3R,4R)-4-mercaptop-3-phenylacetamidoazetidin-2-one (2.31g) and methyl (2RS,5SR)-5-(2-chloroacetyl)tetrahydro-2-furoate (2.02g) in dimethylformamide (30ml). The mixture was stirred at room temperature for 1.5h and then 35 partitioned between ethyl acetate and water. The aqueous phase was separated and extracted with ethyl acetate. The combined organic phases were washed three times with water,

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then brine, dried over magnesium sulphate and evaporated. The title compound (2.208g) was isolated by column chromatography of the residue (silica gel, ethyl acetate as eluent);  $\nu_{\text{max}}$  (CHCl<sub>3</sub>) 3410, 3335, 1777, 1736 and 1678cm<sup>-1</sup>.

5

(d) 4-Methoxybenzyl (RS)-2-hydroxy-2-[4-[(2RS,5SR)-5-methoxycarbonyltetrahydrofuran-2-ylcarbonylmethylthio]-3-phenylacetamidoazetidin-2-on-1-yl]acetate

10 4-Methoxybenzyl glyoxylate hydrate (1.50g) in dichloroethane

(30ml) was heated at reflux for 1h using a Dean and Stark apparatus for heavy entrainers. The mixture was cooled to room temperature and then a solution of (3R,4R)-4-[(2RS,5SR)-5-methoxycarbonyltetrahydrofuran-2-

15 ylcarbonylmethylthio]-3-phenylacetamidoazetidin-2-one

(2.208g) in dichloroethane (20ml) was added followed by triethylamine (0.1ml). The mixture was stirred at room temperature for 1h and then the solvents were evaporated.

The title compound was obtained as a mixture of isomers

20 (2.66g) by column chromatography of the residue using

gradient elution (silica gel, 1:1 hexane:ethyl acetate going to neat ethyl acetate);  $\nu_{\text{max}}$  (CHCl<sub>3</sub>) 3412, 1776, 1741 and 1681cm<sup>-1</sup>.

25 (e) 4-Methoxybenzyl 2-[(3R,4R)-4-[(2RS,5SR)-5-methoxy-

carbonyltetrahydrofuran-2-ylcarbonylmethylthio]-3-

phenylacetamidoazetidin-2-on-1-yl]-2-tri-n-butyl-

phosphoranylideneacetate

30 A solution of thionyl chloride (0.51ml) in tetrahydrofuran

(4ml) was added to a stirred solution of 4-methoxybenzyl

(RS)-2-hydroxy-2-[(2R,4R)-4-[(2RS,5SR)-5-methoxy-

carbonyltetrahydrofuran-2-ylcarbonylmethylthio]-3-phenyl-

acetamidoazetidin-2-on-1-yl]acetate (2.66g) and 2,6-lutidine

35 (0.825ml) in tetrahydrofuran (21ml). The mixture was

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stirred at room temperature for 2h. The solid was filtered off and washed with tetrahydrofuran. The combined filtrates were evaporated and the residue was dissolved in toluene and the solvent evaporated. The residue was dissolved in dioxan 5 (26ml) under argon and then tri-n-butylphosphine (2.6ml) was added. The mixture was stirred at room temperature for 0.5h and then ethyl acetate was added and the solution washed successively with sodium bicarbonate solution, water and brine. The solution was dried over magnesium sulphate and 10 evaporated. The title compound (1.00g) was isolated by column chromatography of the residue using gradient elution (silica gel, 1:1, hexane:ethyl acetate, going to neat ethyl acetate);  $\nu_{\text{max}}$  (CHCl<sub>3</sub>) 3419, 1753, 1676 and 1612cm<sup>-1</sup>.

15 (e) 4-Methoxybenzyl (6R,7R)-3-[(2RS,5SR)-5-methoxy-  
carbonyltetrahydrofuran-2-yl]-7-phenylacetamidoceph-  
3-em-4-carboxylate

A solution of 4-methoxybenzyl 2-[(3R,4R)-4-[(2RS,5SR)-5-  
20 methoxy-carbonyltetrahydrofuran-2-ylcarbonylmethylthio]-3-phenylacetamidoazetidin-2-on-1-yl]-2-tri-n-butyl-phosphoranylideneacetate (1.00g) in toluene (100ml) was heated to reflux for 18h. The solvent was evaporated and the title compound (497mg) separated by column 25 chromatography of the residue using gradient elution (silica gel, 1:1 hexane:ethyl acetate going to neat ethyl acetate);  $\nu_{\text{max}}$  (CHCl<sub>3</sub>) 3409, 1785, 1738 and 1684cm<sup>-1</sup>.

(g) 4-Methoxybenzyl (6R,7R)-7-amino-3-(5-methoxy-  
30 carbonyltetrahydrofuran-2-yl)ceph-3-em-4-carboxylate

A solution of 4-methoxybenzyl (6R,7R)-3-[(2RS,5SR)-5-methoxycarbonyltetrahydrofuran-2-yl]-3-phenylacetamidoceph-3-em-4-carboxylate (497mg) in dichloromethane (7.2ml) was 35 cooled to -15 to -16°C and N-methylmorpholine (0.197ml) was added followed by phosphorus pentachloride in dichloromethane (7.0ml of a solution containing 40mg ml<sup>-1</sup>). The mixture was stirred at the same temperature for 0.5h and

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then methanol (1.8ml) was added and the mixture stirred at room temperature for 0.5h. Water (2.4ml) was added and the mixture vigorously stirred for 0.5h. The dichloromethane was evaporated and the aqueous phase was stirred with ethyl acetate and the pH adjusted to 6.2 with dilute ammonia solution. The organic phase was washed with water, then brine, dried over magnesium sulphate and evaporated. The products were isolated by column chromatography using gradient elution (silica gel, 1:1 hexane:ethyl acetate going to neat ethyl acetate). Eluted first was 4-methoxybenzyl (6R, 7R)-7-amino-3-[(2S, 5R)-5-methoxycarbonyltetrahydrofuran-2-yl]ceph-3-em-4-carboxylate (41mg);  $\nu_{\text{max}}$  (CHCl<sub>3</sub>) 1778 and 1743cm<sup>-1</sup>;  $\delta_{\text{H}}$  (CDCl<sub>3</sub>) 1.6-2.4 (4H, m), 2.68 (2H, br s) 3.65 (1H, d,  $\underline{J}$  18.7Hz), 3.74 (3H, s), 3.80 (3H, s), 3.89 (1H, d,  $\underline{J}$  18.7Hz), 4.49 (1H, dd,  $\underline{J}$  3.2, 8.9Hz), 4.79 (1H, d,  $\underline{J}$  4.7Hz), 4.93 (1H, d,  $\underline{J}$  4.7Hz), 5.06 (1H, dd,  $\underline{J}$  4.9, 9.8Hz), 5.17 (2H, s), 6.89 (2H, d,  $\underline{J}$  8.6Hz) and 7.33 (2H, d,  $\underline{J}$  8.5Hz). Eluted next was 4-methoxybenzyl (6R, 7R)-7-amino-3-[(2R, 5S)-5-methoxycarbonyltetrahydrofuran-2-yl]ceph-3-em-4-carboxylate (126mg);  $\nu_{\text{max}}$  (CHCl<sub>3</sub>) 1777 and 1742cm<sup>-1</sup>;  $\delta_{\text{H}}$  (CDCl<sub>3</sub>) 1.7-2.35 (4H, m), 2.44 (2H, br s), 3.59 (1H, d,  $\underline{J}$  17.8Hz), 3.73 (3H, s), 3.80 (3H, s), 3.98 (1H, d,  $\underline{J}$  17.8Hz), 4.51 (1H, dd,  $\underline{J}$  3.5, 8.8Hz), 4.72 (1H, d,  $\underline{J}$  4.9Hz), 4.94 (1H, d,  $\underline{J}$  4.9Hz), 5.15-5.30 (3H, m), 6.88 (2H, d,  $\underline{J}$  8.7Hz) and 7.34 (2H, d,  $\underline{J}$  8.7Hz).

(h) 4-Methoxybenzyl (6R, 7R)-3-[(2R, 5S)-5-methoxy-carbonyltetrahydrofuran-2-yl]-7-[2-(Z)-methoxyimino-2-(2-tritylaminothiazol-4-yl)acetamido]ceph-3-em-4-carboxylate

30 A stirred solution of 2-(Z)methoxyimino-2-(2-tritylaminothiazol-4-yl)acetic acid hydrochloride (148mg) and N,N-diisopropylethylamine (0.107ml) in dimethylformamide (1ml) was cooled to -55 to -60°C and methanesulphonyl chloride 35 (0.024ml) was added. The mixture was stirred at the same temperature for 0.5h and then a solution of 4-methoxybenzyl

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(6R, 7R)-7-amino-3-[(2R, 5S)-5-methoxy-carbonyltetrahydrofuran-2-yl]ceph-3-em-4-carboxylate (126mg) in dimethylformamide (1ml) was added followed by pyridine (0.023ml). The mixture was then stirred at 0°C for 1h and then at room temperature for 0.5h. The mixture was partitioned between ethyl acetate and aqueous citric acid solution, and the organic phase was washed with water, then brine, dried over magnesium sulphate and evaporated. The title compound (100mg) was isolated by column chromatography of the residue (silica gel, 3:7 hexane:ethyl acetate as eluent);  $\nu_{\text{max}}$  (CHCl<sub>3</sub>) 3403, 1786, 1732 and 1681cm<sup>-1</sup>;  $\delta_{\text{H}}$  (CDCl<sub>3</sub>) 1.66-2.36 (4H, m), 3.58 (1H, d,  $J$  18.0Hz), 3.73 (3H, s), 3.81 (3H, s), 4.02 (1H, d,  $J$  18.0Hz), 4.08 (3H, s), 4.53 (1H, dd,  $J$  3.34, 8.91Hz), 5.02 (1H, d,  $J$  4.8Hz), 5.18 (1H, d,  $J$  11.8Hz), 5.24 (1H, d,  $J$  12.0Hz), 5.30 (1H, dd,  $J$  5.7, 9.9Hz), 5.86 (1H, dd,  $J$  4.6, 8.6Hz), 6.72-6.83 (2H, m), 6.89 (2H, d,  $J$  8.54Hz), 7.01 (1H, s) and 7.25-7.4 (17H, m).

(i) Sodium (6R, 7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(2S, 5S)-5-methoxycarbonyl-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate

Hydrochloric acid (0.12ml of 1N) was added to a stirred solution of 4-methoxybenzyl (6R, 7R)-3-[(2R, 5S)-5-methoxycarbonyltetrahydrofuran-2-yl]-7-[2-(Z)-methoxyimino-2-(2-tritylaminothiazol-4-yl)acetamido]ceph-3-em-4-carboxylate (100mg) in 98% formic acid (2ml). The mixture was stirred at room temperature for 0.5h and then concentrated hydrochloric acid (0.1ml) was added, and the mixture stirred for a further 1h at room temperature. The solid was then filtered off and the filter cake washed with 90% formic acid. The combined filtrates were evaporated and toluene evaporated from the residue twice. The residue was stirred with water and the pH adjusted to 6.2 with saturated aqueous sodium bicarbonate. The solution was filtered and evaporated and the product isolated by column chromatography of the residue (HP20SS using water with increasing

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proportions of acetone as eluent). Fractions containing product were combined, evaporated and the residue dissolved in water (4ml) and freeze-dried to give a mixture (20.7mg) of the title compound;  $\nu_{\text{max}}$  (KBr) 1762, 1669 and  $1603\text{cm}^{-1}$ ;  
5  $\delta_{\text{H}}$  [(CD<sub>3</sub>)<sub>2</sub>SO] 1.54-2.35 (4H, m), 3.23 (1H, d,  $\underline{J}$  17.4Hz),  
3.41 (1H, d,  $\underline{J}$  17.4Hz), 3.63 (3H, s), 3.83 (3H, s), 4.54  
(1H, t,  $\underline{J}$  6.3Hz), 4.97 (1H, d,  $\underline{J}$  4.65Hz), 5.15 (1H, dd,  $\underline{J}$   
5.9, 9.2Hz), 5.55 (1H, dd,  $\underline{J}$  4.6, 7.9Hz), 6.74 (1H, s), 7.23  
(2H, s) and 9.48 (1H, d,  $\underline{J}$  8.1Hz), and the 3-(2R,5S) isomer;  
10  $\delta_{\text{H}}$  (inter alia), 3.66(s), 3.84(s), 4.42 (dd,  $\underline{J}$  3.5, 9.0Hz),  
5.36 (dd,  $\underline{J}$  6.1, 9.7Hz), 6.76(s) and 9.53 (d,  $\underline{J}$  8.3Hz).

(j) 4-Methoxybenzyl (6R,7R)-3-[(2S,5R)-5-methoxy-  
carbonyltetrahydrofuran-2-yl]-7-[2-(2-aminothiazol-4-yl)-2-  
15 (Z)-methoxyiminoacetamido]ceph-3-em-4-carboxylate

A stirred solution of 2-(2-aminothiazol-4-yl)-2-(Z)-  
methoxyiminoacetic acid (20.1mg) and N,N-diisopropyl-  
ethylamine (0.0176ml) in dimethylformamide (0.3ml) was  
20 cooled to -55 to -60°C and methanesulphonyl chloride  
(0.0081ml) was added. The mixture was stirred at the same  
temperature for 0.5h and then a solution of 4-methoxybenzyl  
(6R,7R)-7-amino-3-[(2S,5R)-5-methoxy-  
carbonyltetrahydrofuran-2-yl]ceph-3-em-4-carboxylate (41mg)  
25 in dimethylformamide (0.3ml) was added followed by pyridine  
(0.0073ml). The mixture was then stored at 0°C for 1h and  
then at room temperature for 0.5h. The reaction mixture was  
partitioned between ethyl acetate and aqueous citric acid  
solution and the organic phase washed with water and brine.  
30 The solution was dried over magnesium sulphate and  
evaporated, and the title compound (31mg) isolated by column  
chromatography of the residue (silica gel, ethyl acetate as  
eluent);  $\nu_{\text{max}}$  (CHCl<sub>3</sub>) 3496, 3397, 1784, 1733 and  $1684\text{cm}^{-1}$ .

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(k) Sodium (6R, 7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(2S, 5R)-5-methoxycarbonyltetrahydrofuran-2-yl]ceph-3-em-4-carboxylate

5 A stirred solution of anisole (0.75ml) and dichloromethane (0.38ml) was cooled to -20°C and aluminium chloride (19mg) was added. The mixture was stirred at the same temperature for 15min. and then cooled to -40°C, and then a solution of 4-methoxybenzyl (6R, 7R)-3-[(2S, 5R)-5-methoxycarbonyl-10 tetrahydrofuran-2-yl]-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]ceph-3-em-4-carboxylate (31mg) in dichloromethane (2.5ml) was added and the mixture stirred at the same temperature for 5min. Trisodium citrate (1.64ml of 0.5M solution) was then added and the mixture stirred for 15 10min at room temperature. The aqueous phase was separated and washed twice with dichloromethane. The solution was evaporated and the product isolated by column chromatography of the residue (HP20SS, water with increasing proportions of acetone as eluent). Fractions containing product were 20 combined, evaporated and the residue dissolved in water (3ml) and freeze dried to give the title compound (12mg);  $\nu_{\text{max}}$  (KBr) 1762, 1670 and 1604cm<sup>-1</sup>;  $\delta_{\text{H}}$  [(CD<sub>3</sub>)<sub>2</sub>SO] 1.50-1.63 (1H, m), 1.90-2.26 (3H, m), 3.30-3.47 (2H, m), 3.65 (3H, s), 3.83 (3H, s), 4.39 (1H, dd,  $\text{J}$  3.4, 8.7Hz), 4.98 (1H, d,  $\text{J}$  25 4.8Hz), 5.0 (1H, dd,  $\text{J}$  5.1, 9.8Hz), 5.52 (1H, dd,  $\text{J}$  4.7, 8.2Hz), 6.75 (1H, s), 7.24 (2H, s) and 9.49 (1H, d,  $\text{J}$  8.1Hz).

EXAMPLE 31

30

4-Methoxybenzyl (6R, 7R)-3-(5-acetoxymethyltetrahydrofuran-2-yl)-7-phenylacetamidoceph-3-em-4-carboxylate

35

(a) 5-Acetoxymethylfuran-2-carboxylic acid

A mixture of 5-hydroxymethylfuran-2-carboxylic acid (5.90g), dry dichloromethane (100ml), pyridine (6.71ml), 4-dimethyl-

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aminopyridine (507mg), and acetic anhydride (4.21ml) was stirred for 2 hours at room temperature. The mixture was diluted with ethyl acetate and washed with 5M hydrochloric acid and brine (3 times), dried ( $MgSO_4$ ), and evaporated.

5 The residue was re-evaporated twice from dry toluene to give the title acid as a solid (5.00g);  $\delta_H$  [ $(CD_3)_2CO$ ] 2.05 (3H, s), 5.11 (2H, s), 6.62 (1H, d,  $\underline{\Delta}$  4Hz), 7.17 (1H, d,  $\underline{\Delta}$  4Hz) and 8.31 (1H, br s).

10 (b) (2RS,5SR)-5-Acetoxyethyltetrahydrofuran-2-carboxylic acid

A solution of 5-acetoxyethylfuran-2-carboxylic acid (5.00g) in ethyl acetate (250ml) was stirred with decolourising 15 charcoal (5.0g) for 10mins. The mixture was filtered through Kieselguhr and the residue was washed with ethyl acetate (30ml). The combined filtrates were hydrogenated over 5% rhodium on carbon (2.5g) until hydrogen uptake ceased. The mixture was filtered through Kieselguhr and the 20 residue was washed with ethyl acetate (30ml). The combined filtrates were evaporated to give the title acid as an oil (3.64g);  $\nu_{max}$  (Film) 3700-2800 and  $1742cm^{-1}$ ;  $\delta_H$  [ $(CD_3)_2CO$ ] 1.4-2.5 and 2.00 (7H, m+s), 3.9-4.55 (4H, m) and 7.52 (1H, br s). [Mass spectrum:  $M^+$  (188),  $MH^+$  (189)].

25

(c) (2RS,5SR)-2-Acetoxyethyl-5-bromoacetyltetrahydrofuran

Dry DMF (1 drop) was added to a stirred mixture of the acid from Example 31(b) (500mg) and oxalyl chloride (0.35ml) in 30 dry dichloromethane (10ml). After stirring at room temperature for 1 hour the mixture was evaporated and the residue was re-evaporated from dry dichloromethane (2x2ml) to give the acid chloride as an oil;  $\nu_{max}$  (Film) 1815, 1785 and  $1744cm^{-1}$ .

35

The acid chloride was redissolved in dry dichloromethane (10ml) and treated sequentially with diazomethane (from N-

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methyl-N-nitrosotoluene-4-sulphonamide, 1.65g) and 48% aqueous hydrogen bromide (0.5ml) as for Example 6(a). After stirring at ice bath temperature for 10mins the mixture was washed with water (2x3ml), dried ( $MgSO_4$ ), and evaporated to 5 approximately 5ml to provide a solution of the title bromoketone;  $\nu_{max}$  ( $CH_2Cl_2$ ) 1738cm<sup>-1</sup>.

(d) (3R, 4R)-4-[(2RS, 5SR)-5-Acetoxyethyltetrahydrofuran-2-ylcarbonylmethylthio]-3-phenylacetamidoazetidin-2-one

10 Anhydrous potassium carbonate (183mg) was added portionwise, over 1 minute, to a stirred, ice bath cooled mixture of (3R, 4R)-4-mercapto-3-phenylacetamidoazetidin-2-one (627mg), dry DMF (5ml), and the dichloromethane solution of the 15 bromoketone from Example 31(c). After 15 minutes the cooling bath was removed and the mixture was stirred for an additional 15mins. The mixture was diluted with ethyl acetate (30ml) and was washed with 5% citric acid (5ml), brine (5ml), saturated  $NaHCO_3$  (5ml), and brine (3x5ml). The 20 dried ( $MgSO_4$ ) organic layer was evaporated and the residue was chromatographed on silica gel eluting with ethyl acetate/hexane mixtures and neat ethyl acetate to give the title azetidinones as a gum (495mg);  $\nu_{max}$  ( $CHCl_3$ ) 3411, 3324 br, 1778, 1734 and 1673cm<sup>-1</sup>. [Mass spectrum: +ve ion (3- 25 nitrobenzyl alcohol, sodium acetate)  $MNa^+$  (443)].

(e) 4-Methoxybenzyl (RS)-2-[(3R, 4R)-4-[(2RS, 5SR)-5-acetoxyethyltetrahydrofuran-2-ylcarbonylmethylthio]-3-phenylacetamidoazetidin-2-on-1-yl]-2-hydroxyacetate

30 A mixture of the product from Example 31(d) (490mg), 4-methoxybenzyl glyoxylate monohydrate (272mg), benzene (15ml), and dioxan (2ml) was heated for 1 hour at reflux with provision for the azeotropic removal of water (Dean and

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Stark apparatus containing molecular sieves 4A). The mixture was cooled to room temperature and treated with triethylamine (0.016ml). After stirring at room temperature for 1 hour the mixture was evaporated to give the title compound as a gum;  $\nu_{\text{max}}$  (CHCl<sub>3</sub>) 3613-3159, 1778, 1740 and 1676cm<sup>-1</sup>.

(f) 4-Methoxybenzyl (RS)-2-[(3R,4R)-4-[(2RS,5SR)-5-acetoxyethyltetrahydrofuran-2-ylcarbonylmethylthio]-3-phenylacetamidoazetidin-2-on-1-yl]-2-chloroacetate

The compound from Example 31(e) was dissolved in dry THF (20ml), cooled to -10°C, and treated with 2,6-lutidine (0.20ml) and thionyl chloride (0.13ml). After stirring at 15 -10°C for 10 minutes the mixture was diluted with dry toluene (10ml), filtered, and the residue was washed with dry toluene (10ml). The combined filtrates were evaporated and the residue was re-evaporated from dry toluene (2x3ml) to give the title compound as a gum;  $\nu_{\text{max}}$  (CHCl<sub>3</sub>) 1785, 1742 20 and 1681cm<sup>-1</sup>.

(g) 4-Methoxybenzyl 2-[(3R,4R)-4-[(2RS,5SR)-5-acetoxy-methyltetrahydrofuran-2-ylcarbonylmethylthio]-3-phenylacetamidoazetidin-2-on-1-yl]-2-tri-n-butyl-25 phosphoranylideneacetate

Tri-n-butylphosphine (0.64ml) was added, dropwise over 2 minutes, to a stirred solution of the compound from Example 31(f) in dry dioxan (10ml) at room temperature. After 30 stirring at room temperature for 1 hour the mixture was evaporated and the residue was diluted with ethyl acetate and washed with saturated NaHCO<sub>3</sub> (5ml) and brine (3x5ml). The dried (MgSO<sub>4</sub>) organic layer was evaporated and the residue was chromatographed on silica gel eluting with ethyl 35 acetate/hexane mixtures and neat ethyl acetate to give the title phosphorane as a gum (517mg);  $\nu_{\text{max}}$  (CHCl<sub>3</sub>) 3419, 1749, 1672 and 1611cm<sup>-1</sup>.

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(h) 4-Methoxybenzyl (6R,7R)-3-(5-acetoxymethyl-tetrahydrofuran-2-yl)-7-phenylacetamidoceph-3-em-4-carboxylate

5

A solution of the phosphorane from Example 31(g) (517mg) in dry toluene (100ml) was heated at reflux under dry argon for 8 hours and evaporated. the residue was chromatographed on silica gel eluting with ethyl acetate/hexane mixtures to 10 give two fractions. The less polar fraction contained 4-methoxybenzyl (6R,7R)-3-[(2S,5R)-5-acetoxymethyltetrahydrofuran-2-yl]-7-phenylacetamidoceph-3-em-4-carboxylate, a foam (105mg);  $\nu_{\text{max}}$  (CHCl<sub>3</sub>) 3410, 1784, 1726 and 1683cm<sup>-1</sup>;  $\delta_{\text{H}}$  (CDCl<sub>3</sub>) 1.53-1.83 (3H, m), 2.09 (3H, s), 2.19-2.36 (1H, m), 15 3.30 and 3.55 (2H, ABq,  $J$  18.9Hz), 3.60 and 3.69 (2H, ABq,  $J$  16.2Hz), 3.81 (3H, s), 4.01-4.21 (3H, m), 4.90 (1H, d,  $J$  4.8Hz), 4.96 (1H, dd,  $J$  8.3, 6.7Hz), 5.12 and 5.17 (2H, AA'q,  $J$  12.5Hz), 5.81 (1H, dd,  $J$  9.2, 4.8Hz), 5.97 (1H, d,  $J$  9.2Hz), 6.85-6.92 (2H, m), 7.25-7.41 (7H, m). [Mass 20 spectrum: +ve ion (3-nitrobenzyl alcohol, sodium acetate)  $\text{MH}^+$  (581),  $\text{MNa}^+$  (603)]. The more polar fraction contained 4-methoxybenzyl (6R,7R)-3-[(2R,5S)-5-acetoxymethyl-tetrahydrofuran-2-yl]-7-phenylacetamidoceph-3-em-4-carboxylate, a solid (191mg), m.p. 185-187°C (needles ex 25 ethyl acetate/hexane);  $\nu_{\text{max}}$  (CHCl<sub>3</sub>) 3407, 1785, 1731 and 1682cm<sup>-1</sup>;  $\delta_{\text{H}}$  (CDCl<sub>3</sub>) 1.53-1.78 (2H, m), 1.90-2.05 (2H, m), 2.08 (3H, s), 3.35 and 3.57 (2H, ABq,  $J$  18.0Hz), 3.61 and 3.69 (2H, ABq,  $J$  16.2Hz), 3.80 (3H, s), 4.01-4.19 (3H, m), 4.91 (1H, d,  $J$  4.8Hz), 5.12-5.27 (3H, -m), 5.74 (1H, dd,  $J$  4.8, 9.0Hz), 6.04 (1H, d,  $J$  9.0Hz), 6.85-6.91 (2H, m), 7.25-30 7.41 (7H, m). [Mass spectrum: +ve ion (3-nitrobenzyl alcohol, sodium acetate)  $\text{MH}^+$  (581),  $\text{MNa}^+$  (603)].

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EXAMPLE 32

Sodium (6R,7R)-7-[2-(2-Aminothiazol-4-yl)-2-(2-methoxyiminoacetamido]-3-[3-methyltetrahydrofuran-2-yl]ceph-5-3-em-4-carboxylate

(a) (2RS,3SR)-3-Methyl-2-Tetrahydrofuroic acid

3-Methyl-2-furoic acid (5g) in ethyl acetate (100ml) and 5% 10 rhodium on charcoal catalyst (0.5g) were hydrogenated at ambient temperature and the atmosphere for 6-7h. The catalyst was filtered off and replaced with a further quantity (1g) of catalyst. The reaction mixture was hydrogenated for a further 7h. This procedure was repeated 15 again until no more hydrogen was absorbed. After filtration through kieselguhr and removal of solvent under reduced pressure, the title compound was obtained as a colourless oil (5.096g, quant.);  $\nu_{\text{max}}$  ( $\text{CH}_2\text{Cl}_2$ ) 3674, 3377(br), 1770 and 1722 $\text{cm}^{-1}$ ;  $\delta_{\text{H}}$  ( $\text{CHCl}_3$ ) 1.08 (3H, d,  $\underline{\text{J}}$  7.1Hz), 1.72 (1H, m), 2.16 (1H, m), 2.68 (1H, m), 3.94 (1H, m), 4.18 (1H, m), 4.47 (1H, d,  $\underline{\text{J}}$  7.5Hz) and 9.42 (1H, v. br s, exch). [Mass spectrum: +ve ion (ammonia)  $\text{MNH}_4^+$  (148)].

(b) (2RS,3SR)-2-Bromoacetyl-3-Methyltetrahydrofuran

25 (2RS,3SR)-3-Methyl-2-tetrahydrofuroic acid 91.3g) was converted to the acid chloride with oxalyl chloride (2.54g, 1.75mls) in dichloromethane (20mls) as described in Example 1(a). Diazomethane was passed through a solution of the 30 acid chloride in dichloromethane (20mls), cooled in ice/water until i.r. analysis showed no starting material. Hydrobromic acid (2mls, 49% w/v aqueous solution), was added dropwise and the reaction mixture stirred vigorously for 10min. T.l.c. analysis showed complete conversion to the 35 title compound. The solution was washed with water, brine and dried. The solvent was washed with water, brine and

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dried. The solvent was evaporated and the residue flash chromatographed on silica gel, eluting with 5 and then 10% ethyl acetate/hexane to give the product as an almost colourless oil, (1.621g, 79%);  $\nu_{\text{max}}$  ( $\text{CH}_2\text{Cl}_2$ )  $1732\text{cm}^{-1}$ ;  $\delta_{\text{H}}$  5 ( $\text{CDCl}_3$ ) 0.96 (3H, d,  $\underline{\text{J}}$  7.2Hz), 1.70 (1H, m), 2.17 (1H, m), 2.70 (1H, m), 3.93 (1H, m), 4.12 and 4.25 (2H, ABq,  $\underline{\text{J}}$  14.7Hz) and 4.49 (1H, d,  $\underline{\text{J}}$  7.3Hz). [Mass spectrum: +ve ion (ammonia)  $\text{MNH}_4$  (224)].

10 (c) 4-Methoxybenzyl (2RS)-2-Hydroxy-2-[(3R,4R)-3-phenylacetamido-4-[(2RS,3SR)-3-methyltetrahydrofuran-2-ylcarbonylmethylthio]azetidin-2-on-1-yl]acetate

4-Methoxybenzyl (2RS)-2-hydroxy-2-[(1R,5R)-3-benzyl-4-thia-15 2,6-diazabicyclo[3.2.0]hept-2-en-7-on-6-yl]acetate (12.66g) was hydrolysed in 50% dichloromethane-acetone (80ml) with toluene-4-sulphonic acid hydrate (10.22g) in water (25ml) as described in Example 6(b). The crude thiol thus prepared (12.942g), in acetone (50ml) was treated with (2RS,3SR)-2-20 bromoacetyl-3-methyltetrahydrofuran (6.57g) in acetone (20ml) for 10min. at room temperature. Then potassium carbonate (2.08g) was added and stirring continued for 30min. The solution was diluted with ethyl acetate (200ml), washed with water (2x), brine and then dried. Removal of 25 solvent gave a yellow gum. Flash chromatography on silica gel eluting with 50, 60, 70, 80 and then 90% ethyl acetate-hexane afforded the title compound as a pale yellow foam (10.406g, 62%);  $\nu_{\text{max}}$  ( $\text{CH}_2\text{Cl}_2$ ) 3405(br), 1780, 1744, 1683 and 1613 $\text{cm}^{-1}$ .

30

(d) 4-Methoxybenzyl 2-[(3R,4R)-3-Phenylacetamido-4-[(2RS,3SR)-3-methyltetrahydrofuran-2-ylcarbonylmethylthio]azetidin-2-on-1-yl]-2-tri-n-butylphosphoranylideneacetate

35

4-Methoxybenzyl (2RS)-2-hydroxy-2-[(3R,4R)-3-phenylacetamido-4-[(2RS,3SR)-3-methyltetrahydrofuran-2-ylcarbonylmethylthio]azetidin-2-on-1-yl]acetate (10.406g)

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was converted to it's chloride with thionyl chloride (3.34g, 2.02ml) and 2,6-lutidine (3.00g, 3.25ml) in tetrahydrofuran (100ml) as described in Example 6(c). The crude chloride in dioxan (80ml) was then converted to the product with tri-n-<sub>5</sub> butylphosphine (6.98ml) also described in 6(c). Flash chromatography on silica gel afforded the title compound as a foam (6.525g, 47%). [Mass spectrum: +ve ion (3-nitrobenzyl alcohol, sodium acetate)  $\text{MNa}^+$  (763)].

10 (e) 4-Methoxybenzyl (6R,7R)-7-Phenylacetamido-3-[  
1(2RS,3SR)-3-methyltetrahydrofuran-2-yl]ceph-3-em-4-  
carboxylate

The phosphorane from Example 32(d), (6.525g) in xylene (120ml) heated under reflux for 6-7h. until t.l.c. analysis (ethyl acetate) showed no more starting material. Concentration and flash chromatography on silica gel eluting with 30 and then 40% ethyl acetate in hexane gave the diastereoisomer mixture of the product as a brown foam (1.293g, 28%); The <sup>1</sup>H n.m.r. spectrum showed substantial amounts of the  $\Delta$ -2 isomeric cephems. The crude mixture in methanol (15ml) and dichloromethane (5ml) was treated at room temperature with a solution of sodium metaperiodate (0.636g) in water (5ml) overnight and then heated to about 25 60°C for 1h. The precipitate was filtered off and the filtrate concentrated. The residue was partitioned between ethyl acetate-water. The organic phase was then dried and concentrated. The residual gum was purified by flash chromatography on silica gel, eluting with 50%, 70% ethyl acetate-hexane and then neat ethyl acetate. The sulphoxide derivative of the cephem was obtained as a yellow foam, (0.484g, 35%). This foam was dissolved in dimethylformamide (5ml), cooled, under argon, to -30°C. Phosphorus trichloride (0.239g, 0.152ml) was added and the solution 35 stirred for ca. 1h. The solution was then diluted with ethyl acetate and washed with water (3x) and then brine. After drying and removal of solvent the crude title compound was obtained as a brown foam, (0.441g, 97%); a sample of the

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crude product was flash chromatographed on silica gel, eluting with 40%, 50% ethyl acetate-hexane and afforded the less polar isomer as a pale yellow foam;  $\nu_{\text{max}}$  (CH<sub>2</sub>Cl<sub>2</sub>) 3415, 1783, 1722, 1688 and 1613cm<sup>-1</sup>;  $\delta_{\text{H}}$  (CDCl<sub>3</sub>) 0.76 (3H, d,  $J$  7.2Hz), 1.55 (1H, m), 2.14 (1H, m), 2.73 (1H, m), 3.26 and 3.55 (2H, ABq,  $J$  18.6Hz), 3.60-3.77 (3H, m), 3.82 (3H, s), 4.04 (1H, m), 4.90 (1H, d,  $J$  4.8Hz), 4.93 (1H, d,  $J$  7.6Hz), 5.16 (2H, s), 5.80 (1H, dd,  $J$  4.8, 9.0Hz), 6.04 (1H, d,  $J$  9.0Hz), 6.88 (2H, m) and 7.24-7.41 (7H, m). [Mass spectrum: 10 +ve ion (3-nitrobenzyl alcohol, sodium acetate),  $\text{MNa}^+$  (545)]. The second, more polar isomer was then eluted and isolated as a pale yellow solid;  $\nu_{\text{max}}$  (CH<sub>2</sub>Cl<sub>2</sub>) 3414, 1782, 1726, 1688 and 1613cm<sup>-1</sup>;  $\delta_{\text{H}}$  (CDCl<sub>3</sub>) 0.79 (1H, d,  $J$  7.2Hz), 1.53 (1H, m), 2.07 (1H, m), 2.40 (1H, m), 3.24 and 3.49 (2H, ABq,  $J$  18.1Hz), 3.57-3.75 (3H, m), 3.82 (3H, s), 3.91 (1H, m), 4.93 (1H, d,  $J$  4.7Hz), 5.18 (2H, s), 5.26 (1H, d,  $J$  6.7Hz), 5.71 (1H, dd,  $J$  4.7, 9.0Hz), 6.08 (1H, d,  $J$  9.0Hz), 6.88 (2H, d,  $J$  8.7Hz) and 7.26-7.40 (7H, m). [Mass spectrum: +ve ion (3-nitrobenzyl alcohol, sodium acetate), 15  $\text{MNa}^+$  (545)].

(f) 4-Methoxybenzyl (6R,7R)-7-Amino-3-[(2RS,3SR)-3-methyltetrahydrofuran-2-yl]ceph-3-em-4-carboxylate

25 4-Methoxybenzyl (6R,7R)-7-phenylacetamido-3-[(2RS,3SR)-3-methyltetrahydrofuran-2-yl]ceph-3-em-4-carboxylate (0.692g) in dry dichloromethane (5ml) under argon was cooled to -20°C. This solution was then treated with 4-methylmorpholine (0.268g, 0.291ml) followed by a solution of 30 phosphorus pentachloride in dichloromethane (0.415g in 10.37ml) in a rapid dropwise fashion. The solution was allowed to warm to -5°C and maintained at this temperature for 0.5h. Methanol (5ml) was then added in one portion and the solution allowed to warm to room temperature, and 35 stirred for 0.5h. Water (5ml) was then added and the solution rapidly stirred for a further 0.75h. The dichloromethane was evaporated at reduced pressure and replaced with ethyl acetate. The pH was adjusted to 7.5

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with aqueous 880 ammonia. The aqueous phase was extracted with ethyl acetate and the combined organic layers washed with brine and dried. Removal of solvent and column chromatography on silica gel eluting with 60 and then-70% 5 ethyl acetate in hexane afforded the (2S,3R) isomer of the title compound as a pale yellow foam, (0.197g, 37%);  $\nu_{\text{max}}$  ( $\text{CH}_2\text{Cl}_2$ ) 1777, 1720 and  $1613\text{cm}^{-1}$ ;  $\delta_{\text{H}}$  ( $\text{CDCl}_3$ ) 0.89 (3H, d,  $J$  7.2Hz), 1.56 (1H, m), 2.14 (1H, m), 2.73 (1H, m), 3.37 and 3.57 (2H, ABq,  $J$  18.0Hz), 3.73 (1H, m), 3.82 (3H, s), 4.05 10 (1H, m), 4.79 (1H, d,  $J$  4.8Hz), 4.93 (1H, d, 4.8Hz), 4.98 (1H, d,  $J$  7.6Hz), 5.17 (2H, s), 6.88 (2H, d,  $J$  8.6Hz) and 7.33 (2H, d,  $J$  8.6Hz). [Mass spectrum: +ve ion (3-nitrobenzyl alcohol, sodium acetate)  $\text{MH}^+$  (405),  $\text{MNa}^+$  (427)].

15 The second compound to be eluted was the (2R,3S) isomer of the title compound, as a brown gum, (0.193g, 36%);  $\nu_{\text{max}}$  ( $\text{CH}_2\text{Cl}_2$ ) 1775, 1727 and  $1613\text{cm}^{-1}$ ;  $\delta_{\text{H}}$  ( $\text{CDCl}_3$ ) 0.84 (3H, d,  $J$  7.0Hz), 1.54 (1H, m), 2.06 (1H, m), 2.45 (1H, m), 3.33 and 3.64 (2H, ABq,  $J$  17.5Hz), 3.71 (1H, m), 3.82 (3H, s), 3.92 20 (1H, m), 4.78 (1H, d,  $J$  4.5Hz), 4.98 (1H, d,  $J$  4.5Hz), 5.18 (2H, s), 5.29 (1H, d,  $J$  8.1Hz), 6.88 (2H, d,  $J$  8.6Hz) and 7.33 (2H, d,  $J$  8.6 Hz). [Mass spectrum: +ve ion (3-nitrobenzyl alcohol, sodium acetate)  $\text{MH}^+$  (405),  $\text{MNa}^+$  (427)]. Also isolated was a mixture of the isomers, (0.083g, 15%).

25

(g) 4-Methoxybenzyl (6R,7R)-7-[2-(2-Aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(2S,3R)-3-methyltetrahydrofuran-2-yl]ceph-3-em-4-carboxylate

30 Methanesulphonyl chloride (0.04ml) was added to 2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetic acid (0.103g) and N,N-diisopropylethylamine (0.089ml) in DMF (1ml) under argon at  $-50^{\circ}\text{C}$ . The solution was maintained between  $-30^{\circ}\text{C}$  and  $-40^{\circ}\text{C}$  for 1h. A solution of the (2S,3R)-isomer from 35 Example 34(f), (0.188g) and pyridine (0.038ml) in DMF (1ml) was added and the solution warmed to room temperature over

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1h. The reaction mixture was diluted with ethyl acetate, washed successively with saturated sodium hydrogencarbonate, water, brine and then dried. After removal of solvent under vacuum, the residue was flash chromatographed on silica gel, eluting with 50, 70, 80 and then 90% ethyl acetate-hexane to give the title compound as a waxy solid, (0.227g, 83%);  $\nu_{\text{max}}$  ( $\text{CH}_2\text{Cl}_2$ ) 3482, 3389, 1783, 1722, 1688, 1613 and  $1516\text{cm}^{-1}$ ;  $\delta_{\text{H}}$  ( $\text{CDCl}_3$ ) 0.89 (3H, d,  $\underline{J}$  7.1Hz), 1.57 (1H, m), 2.15 (1H, m), 2.76 (1H, m), 3.35 and 3.61 (2H, ABq,  $\underline{J}$  18.6Hz), 3.74 (1H, m), 3.82 (3H, s), 4.04 (1H, m), 4.10 (3H, s), 4.98 (1H, d,  $\underline{J}$  7.6Hz), 5.04 (1H, d,  $\underline{J}$  4.8Hz), 5.19 (2H, s), 5.22 (2H, br s, exch.), 5.94 (1H, dd,  $\underline{J}$  4.8, 8.9Hz, collapses to d,  $\underline{J}$  4.8Hz on exch.), 6.91 (2H, d,  $\underline{J}$  8.6Hz), 6.99 (1H, s), 7.20 (1H, d,  $\underline{J}$  8.9Hz, exch.) and 7.34 (1H, d,  $\underline{J}$  8.6Hz). [Mass spectrum: 15 +ve ion (3-nitrobenzyl alcohol, sodium acetate)  $\underline{M}\text{H}^+$  (588),  $\underline{M}\text{Na}^+$  (610)].

(h) 4-Methoxybenzyl (6R,7R)-7-[2-(2-Aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(2R,3S)-3-methyltetra-20 hydrofuran-2-yl]ceph-3-em-4-carboxylate

The procedure used in Example 32(g) was repeated for the (2R,3S) isomer from Examaple 32(h), (0.183g); with 2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetic acid (0.1g), 25 N,N-diisopropylethylamine (0.087ml), methanesulphonyl chloride (0.039ml) and pyridine (0.037ml). After work up and purification the title compound was obtained as a pale yellow foam, (0.185g, 70%);  $\nu_{\text{max}}$  ( $\text{CH}_2\text{Cl}_2$ ) 3484, 3388, 1782, 1731, 1688, 1609 and  $1516\text{cm}^{-1}$ ;  $\delta_{\text{H}}$  ( $\text{CDCl}_3$ ) 0.84 (2H, d,  $\underline{J}$  7.1hz), 1.56 (1H, m), 2.09 (1H, m), 2.46 (1H, m), 3.33 and 3.60 (2H, ABq,  $\underline{J}$  17.8Hz), 3.73 (1H, m), 3.83 (3H, s), 3.93 (1H, m), 4.09 (3H, s), 5.07 (1H, d,  $\underline{J}$  4.6Hz), 5.21 (2H, s), 5.30 (1H, d,  $\underline{J}$  6.9Hz), 5.02 (2H, br s, exch.), 5.87 (1H, dd  $\underline{J}$  4.6, 8.7Hz collapses to d,  $\underline{J}$  4.6Hz on exch.), 6.90 (2H, d,  $\underline{J}$  8.7Hz), 6.98 (1H, s) and 7.35 (3H, d,  $\underline{J}$  8.7Hz overlapping m, exch.). [Mass spectrum: +ve ion (3-nitrobenzyl alcohol, sodium acetate)  $\underline{M}\text{N}$  (588),  $\underline{M}\text{Na}^+$  (610)].

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(i) Sodium (6R,7R)-7-[2-(2-Aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(2S,3R)-3-methyltetrahydrofuran-2-yl]ceph-3-em-4-carboxylate

5 A mixture of dichloromethane (3ml) and anisole (6ml) under argon was cooled to -20°C and aluminium trichloride (0.15g) was added. The solution was stirred for 0.25h and then cooled to -40°C. A solution of the cephem prepared in Example 32(g) in dichloromethane (6ml) was added in one 10 portion. T.l.c. analysis (ethyl acetate) immediately after addition showed no starting material. A solution of trisodium citrate (12ml, 0.5M solution) was added and the mixture vigorously stirred for 10 minutes at room 15 temperature. The aqueous phase was separated, washed twice with dichloromethane and concentrated to about 5ml. Column chromatography on HP20SS eluting with 0, 1, 2 and 4% tetrahydrofuran in water, followed by concentration and freeze-drying of the relevant fractions afforded the title compound as an amorphous white solid, (0.134g, 73%);  $\nu_{\text{max}}$  20 (KBr) 1761, 1667, 1597 and 1531cm<sup>-1</sup>;  $\delta$  ( $d_6$ -DMSO) 0.86 (3H, d,  $\underline{J}$  7.1Hz), 1.48 (1H, m), 2.00 (1H, m), 2.56 (1H, m), 3.09 and 3.37 (2H, ABq,  $\underline{J}$  17.1Hz), 3.58 (1H, m), 3.84 (3H, s), 3.41 (1H, m), 4.92 (1H, d,  $\underline{J}$  7.7Hz), 4.96 (1H, d,  $\underline{J}$  4.6Hz), 5.53 (1H, dd,  $\underline{J}$  4.6, 8.1Hz, collapses to d,  $\underline{J}$  4.6Hz on 25 exch.), 6.74 (1H, s), 7.24 (2H, br s, exch.) and 9.57 (1H, d,  $\underline{J}$  8.1Hz, exch.). [Mass spectrum: +ve ion (thioglycerol)  $\text{MH}^+$  (490),  $\text{MNa}^+$  (512)].

(j) Sodium (6R,7R)-7-[2-(2-Aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(2R,3S)-3-methyltetrahydrofuran-2-yl]ceph-3-em-4-carboxylate

The procedure used for Example 32(i) with dichloromethane (2.5ml), anisole (5ml), aluminium trichloride (0.12g) and 35 the (2R,3S) isomer (0.178g) was employed. Following work up with trisodium citrate (10ml, 0.5M solution) the product was

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isolated and purified as described to give the title compound as an amorphous white solid, (0.117g, 79%);  $\nu_{\text{max}}$  (KBr) 1762, 1665, 1597, 1532 and 1456cm<sup>-1</sup>;  $\delta_{\text{H}}$  ( $\text{d}_6$ -DMSO) 0.86 (3H, d, J 7.0Hz), 1.47 (1H, m), 2.00 (1H, m), 2.28 (1H, m), 3.18 and 3.38 (2H, ABq, J 16.9Hz), 3.58 (1H, m), 3.86 (3H, s), 3.93 (1H, m), 4.99 (1H, d, J 4.5Hz), 5.44 (1H, d, J 7.7Hz), 5.50 (1H, dd, J 4.5, 8.6Hz, collapses to d, J 4.5Hz on exch.), 6.76 (1H, s), 7.25 (2H, br s, exch.) and 9.50 (1H, d, J 8.6Hz, exch.). [Mass spectrum: +ve ion (thioglycerol)  $\text{MH}^+$  (490),  $\text{MNa}^+$  (512)].

EXAMPLE 33

4-Methoxybenzyl (6R,7R)-3-[tetrahydropyran-4-yl]-7-phenylacetamidoceph-3-em-4-carboxylate

(a) 4-Methoxybenzyl (2RS)-2-hydroxy-2-[(3R,4R)-3-phenylacetamido-4-(tetrahydropyran-4-ylcarbonyl-methylthio)azetidin-2-on-1-yl]acetate

Crude 4-methoxybenzyl (2RS)-2-hydroxy-2-[(3R,4R)-4-mercaptop-3-phenylacetamidoazetidin-2-on-1-yl]acetate (prepared from 4-methoxybenzyl (2RS)-2-hydroxy-2-[(1R,5R)-3-benzyl-4-thia-2,6-diazabicyclo[3.2.0]hept-2-en-7-on-6-yl]acetate (8.35g, 20mmol)) was dissolved in acetone (25ml) and treated with a solution of 4-bromoacetyltetrahydropyran (G.H. Harnest and A. Burger, J. Amer. Chem. Soc., 1943, 65, 370) (4.4g, 20mmol). After 20min., potassium carbonate (1.38g, 10mmol) was added and the mixture stirred again for a further 45min. Excess ethyl acetate was then added and the organic solution washed with water, brine and dried over anhydrous  $\text{MgSO}_4$ . Evaporation of solvent and chromatography of the residue on silica gel using 50% hexane in ethyl acetate to 100% ethyl acetate gave the title compound as a pale yellow foam (8.5g; 76%);  $\nu_{\text{max}}$  ( $\text{CHCl}_3$ ) 3420, 1780, 1750, 1680 and 1615cm<sup>-1</sup>. [Mass spectrum: +ve ion (3-nitrobenzyl alcohol, sodium acetate)  $\text{MNa}^+$  (579)].

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(b) 4-Methoxybenzyl 2-[(3R,4R)-3-phenylacetamido-4-[tetrahydropyran-4-ylcarbonylmethylthio]azetidin-2-on-1-yl]-2-tri-n-butylphosphoranylidene acetate

5 A solution of thionylchloride (1.ml, 15mmole) in THF (10ml) was added dropwise to the hydroxy compound from Example 33(a) (5.56, 10mmol) and 2,6-lutidine (1.75ml, 15mmol) in THF (30ml) at -20°C. After stirring for 30min. the reaction was filtered through a pad of celite and the filtrate 10 evaporated. Toluene was added and re-evaporated to yield 4-methoxybenzyl (RS)-2-chloro-2-[(3R,4R)-3-phenylacetamido-4-[tetrahydropyran-4-ylcarbonylmethylthio]azetidin-2-on-1-yl]acetate as a dark brown oil

15 The crude chloro compound was dissolved in dioxan (30ml) and treated with tri-n-butylphosphine (5.5ml, 22mmol). After stirring for 1h. at room temperature the reaction mixture was diluted with ethyl acetate and washed successively with dilute aqueous sodium bicarbonate solution, water and brine.

20 After drying over anhydrous magnesium sulphate the solvent was evaporated. Chromatography of the residue on silica gel using ethyl acetate as eluent gave the title compound as a brown foam (6.2g, 84%);  $\nu_{\text{max}}$  (CHCl<sub>3</sub>) 3450, 1760, 1675, 1615 and 1510cm<sup>-1</sup>. [Mass spectrum: +ve ion (3-nitrobenzyl 25 alcohol, sodium acetate)  $\text{MH}^+$  (741)  $\text{MNa}^+$  (763)].

(c) 4-Methoxybenzyl (6R,7R)-7-phenylacetamido-3-[tetrahydropyran-4-yl]ceph-3-em-4-carboxylate

30 A solution of the phosphorane from Example 33(b) (6g) and benzoic acid (20mg) in xylene (500ml) was heated at reflux for 44h. The reaction mixture was cooled, concentrated and the residue purified by chromatography on silica gel with 50% ethyl acetate in hexane to give the title compound as a

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mixture with the  $\Delta^2$  cephem (1:2) (1.24g);  $\nu_{\text{max}}$  (CHCl<sub>3</sub>) 3420, 1780, 1730, 1680 and 1615cm<sup>-1</sup>;  $\delta_{\text{H}}$  (CDCl<sub>3</sub>),  $\Delta^3$  isomer, 1.20-1.90 (4H, m), 2.05-2.20 (1H, m), 3.10-3.40 (4H, m), 3.62 and 3.67 (2H, ABq,  $J$  16.0Hz), 3.81 (3H, s), 3.85-4.05 (2H, m), 5 4.90 (1H, d,  $J$  4.7Hz), 5.13 and 5.24 (2H, ABq,  $J$  11.8Hz), 5.77 (1H, dd,  $J$  4.7, 9.1Hz), 6.00 (1H, d,  $J$  9.1Hz), 6.89 (2H, d,  $J$  8.6Hz) and 7.25-7.45 (7H, m). [Mass spectrum: +ve ion (ammonia) 523 (MH<sup>+</sup>), 540 (MNH<sub>4</sub><sup>+</sup>)].

10 EXAMPLE 344-Methoxybenzyl (6R,7R)-3-[(2R,3R,4S)-3,4-dimethoxy-tetrahydrofuran-2-yl]-7-phenylacetamidoceph-3-em-4-carboxylate

15

(a) 1,4-Anhydro-2,3-O,O-dimethyl-5,6-O-isopropylidene-D-glucitol

1,4-Anhydro-5,6-O-isopropylidene-D-glucitol (S. Soltzberg, 20 R.M. Goepp, Jr., and W. Freudenberg, J. Amer. Chem. Soc., 1946, 68, 919) (8.74g, 43mmol), methyl iodide (11ml, 172mmol) and silver oxide (29.9g, 129mmol) in DMF (50ml) were stirred overnight, filtered through celite and evaporated in vacuo. The residue was extracted with ether, 25 filtered through celite and evaporated to give the title compound as a colourless oil (8.26g, 83%);  $\nu_{\text{max}}$  (CH<sub>2</sub>Cl<sub>2</sub>) 1675, 1457, 1381, 1270, 1216, 1108 and 1073cm<sup>-1</sup>;  $\delta_{\text{H}}$  (CDCl<sub>3</sub>, 250MHz) 1.37 (3H, s), 1.43 (3H, s), 3.38 (3H, s), 3.45 (3H, s), 3.7-4.35 (8H, m).

30

(b) 1,4-Anhydro-2,3-O,O-dimethyl-D-glucitol

The product from Example 34(a) (8.26g) in ethanol (32ml) and water (8ml) was stirred with Amberlite IR 120 (H<sup>+</sup>) (20g moist) for 4h. then filtered and evaporated to dryness to 35 provide the title compound as an oil (6.50g, 95%);  $\nu_{\text{max}}$  (CH<sub>2</sub>Cl<sub>2</sub>) 3583, 3460, 1462, 1108, 1179 and 1061cm<sup>-1</sup>;  $\delta_{\text{H}}$

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(CDCl<sub>3</sub>, 250MHz) 2.13 (br s, exch.), 3.39 (3H, s), 3.47 (3H, s), 3.65-4.0 (7H, m), 4.09 (1H, dd,  $\underline{\Delta}$  4.63, 9.87Hz). [Mass spectrum: +ve ion (ammonia)  $\underline{\text{MNH}_4}^+$  (193),  $\underline{\text{MNH}_4}^+$  (210)].

5 (c) (2S,3R,4S)-3,4-Dimethoxytetrahydrofuran-2-yl-carboxaldehyde

Sodium metaperiodate (7.97g, 37mmol) in water (50ml) was added to an ice bath cooled solution of 1,4-anhydro-2,3-O,O-10 dimethyl-D-glucitol (6.50g, 34mmol) in methanol (150ml) and then mixture stirred 0.5h then filtered and the filtrate evaporated in vacuo. The residue was extracted five times with dichloromethane then the combined extracts were dried (MgSO<sub>4</sub>) and evaporated to give the crude aldehyde as a 15 colourless oil (5.734g);  $\nu_{\text{max}}$  (CH<sub>2</sub>Cl<sub>2</sub>) 3445, 1735, 1463, 1194 and 1120cm<sup>-1</sup>;  $\delta_{\text{H}}$  (CDCl<sub>3</sub>, 250MHz), 3.38 (3H, s), 3.39 (3H, s), 3.94 (1H, d,  $\underline{\Delta}$  3.87Hz), 4.02 (1H, d,  $\underline{\Delta}$  9.99Hz), 4.14 (1H, d,  $\underline{\Delta}$  4.77Hz), 4.20 (1H, dd,  $\underline{\Delta}$  3.90, 10.07Hz), 4.39 (1H, dd,  $\underline{\Delta}$  1.77, 4.72Hz) and 9.65 (1H, d,  $\underline{\Delta}$  1.80Hz). [Mass 20 spectrum: +ve ion (ammonia)  $\underline{\text{MNH}_4}^+$  (178)].

(d) (2S,3R,4S)-3,4-Dimethoxytetrahydrofuran-2-ylcarboxylic acid

25 Jones reagent (R.G. Curtis, I. Heilbron, E.R.H. Jones and G.F. Woods, J. Chem. Soc., 1953, 457) (11ml) was added dropwise to the aldehyde (5.73g) from Example 34(c) in acetone (125ml) cooled in an ice bath. After 10 minutes the orange solution was treated with propan-2-ol (2ml), stirred 30 a further 10 minutes then diluted with ether (125ml), filtered through celite and evaporated in vacuo. The residue in dichloromethane was dried (MgSO<sub>4</sub>), concentrated and flash chromatographed on silica gel eluting with 60, 70 and 80% ethyl acetate in hexane to give the title compound 35 (4.68g, 72%) as a colourless oil;  $\nu_{\text{max}}$  (CH<sub>2</sub>Cl<sub>2</sub>) 3404(br), 1760, 1735, 1462, 1368, 1113, 1094 and 1056cm<sup>-1</sup>;  $\delta_{\text{H}}$  (CDCl<sub>3</sub>, 250MHz) 3.40 (3H, s), 3.44 (3H, s), 3.94 (1H, d,  $\underline{\Delta}$  3.83Hz), 4.00 (1H, d,  $\underline{\Delta}$  9.90Hz), 4.07 (1H, d,  $\underline{\Delta}$  4.16Hz) and 4.21 (1H,

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dd,  $J$  3.83, 9.85Hz). [Mass spectrum: +ve ion (ammonia)  $\underline{\text{MNH}_4}^+$  (194)].

(e) (2S,3R,4S)-2-Bromoacetyl-3,4-dimethoxytetrahydrofuran

5

A solution of (2S,3R,4S)-3,4-dimethoxytetrahydrofuran-2-carboxylic acid (3.0g, 17.0mmol) in dichloromethane (30ml) was treated with oxalyl chloride (3.0ml, 34.4mmol) and dimethylformamide (3 drops). The mixture was stirred for 10 1h., evaporated in vacuo, dichloromethane added, and re-evaporated. The resulting acid chloride was dissolved in dichloromethane (30ml) and cooled in an ice-bath. Diazomethane was then passed into the solution as described in Example 14(a). When the addition was complete, 48% aqueous hydrogen bromide (3.2ml) was added, and the mixture stirred for a further 10min. The solution was washed with water (x2), dried over  $\text{MgSO}_4$  and concentrated in vacuo to yield the title compound (3.40g, 79%); (Found:  $\underline{\text{M}}^+$  251.9986.  $\text{C}_3\text{H}_13\text{O}_4\text{Br}$  requires 251.9997);  $\nu_{\text{max}}$  ( $\text{CH}_2\text{Cl}_2$ ) 1739 $\text{cm}^{-1}$ ;  $\delta_{\text{H}}$  (CDCl<sub>3</sub>, 250MHz) 3.37 (3H, s), 3.39 (3H, s) and 3.41-4.70 (7H, series of m).

(f) 4-Methoxybenzyl (2RS)-2-hydroxy-2-[(3R,4R)-4-[(2S,3R,4S)-3,4-dimethoxytetrahydrofuran-2-ylcarbonyl-methylthio]-3-phenylacetamidoazetidin-2-on-1-yl]acetate

4-Methoxybenzyl (RS)-2-hydroxy-2-[(1R,5R)-3-benzyl-4-thia-2,6-diazabicyclo[3.2.0]hept-2-en-7-on-6-yl]acetate (6.0g, 14.6mmol) in 50% acetone/dichloromethane (60ml) was cleaved 30 with 4-toluenesulphonic acid (5.0g, 26.3mmol) in water (12ml). The product was reacted with crude bromide from Example 34(e) (3.40g, 13.4mmol) in acetone (70ml) followed by potassium carbonate (1.0g, 7.2mmol) as described in Example 6(b). After work-up, the residue was purified by 35 chromatography on silica gel eluting with 50, 70 and 100% ethyl acetate in hexane to yield the title compound (3.40g,

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42%);  $\nu_{\text{max}}$  ( $\text{CH}_2\text{Cl}_2$ ) 3400, 1781, 1735, 1682, 1613 and 1516 $\text{cm}^{-1}$ . [Mass spectrum: +ve ion (3-nitrobenzyl alcohol, sodium acetate)  $\text{MNa}^+$  (625)].

5 (g) 4-Methoxybenzyl 2-[(3R,4R)-4-[(2S,3R,4S)-3,4-dimethoxytetrahydrofuran-2-ylcarbonylmethylthio]-3-phenylacetamidoazetidin-2-on-1-yl]-2-tri-n-butyl-phosphoranylideneacetate

10 The alcohol from Example 34(f) (3.35g, 5.56mmol) was treated with thionyl chloride (623 $\mu\text{l}$ , 8.54mmol) and 2,6-lutidine (995 $\mu\text{l}$ , 8.54mmol), followed by tri-n-butylphosphine (3.12ml, 12.52mmol) as described for Example 6(c). The product was purified by chromatography on silica gel eluting with 0 and 15 10% methanol in ethyl acetate to yield the title compound (2.68g, 61%);  $\nu_{\text{max}}$  ( $\text{CH}_2\text{Cl}_2$ ) 1761, 1682, 1613 and 1515 $\text{cm}^{-1}$ . [Mass spectrum: +ve ion (3-nitrobenzyl alcohol, sodium acetate)  $\text{MH}^+$  (787),  $\text{MNa}^+$  (809)].

20 (h) 4-Methoxybenzyl (6R,7R)-3-[(2R,3R,4S)-3,4-dimethoxytetrahydrofuran-2-yl]-7-phenylacetamidoceph-3-em-4-carboxylate

A solution of the phosphorane from Example 34(g) (2.60g, 25 3.30mmol) and benzoic acid (10mg) in toluene (40ml) was

heated to reflux for 16h. After cooling, the solvent was evaporated in vacuo. The residue was purified by chromatography on silica gel eluting with 10, 30 and 50% ethyl acetate in hexane to yield the title compound

30 contaminated with the  $\Delta$ 2-isomer (296mg, 16%);  $\nu_{\text{max}}$  ( $\text{CH}_2\text{Cl}_2$ ) 3418, 1783, 1732, 1682, 1612 and 1515 $\text{cm}^{-1}$ ;  $\delta_{\text{H}}$ ,  $\Delta$ 3-isomer ( $\text{CDCl}_3$ , 250MHz) 3.25 (3H, s), 3.31 (3H, s), 3.32-4.16 (8H, series of m), 3.80 (3H, s), 4.92 (1H, d,  $J$  4.8Hz), 4.98-5.28 (3H, m), 5.77 (1H, dd,  $J$  9.2, 4.8Hz), 6.00 (1H, br d,  $J$  35 9.2Hz, exch.), 6.88 (2H, d,  $J$  8.6Hz) and 7.22-7.41 (7H, m). [Mass spectrum:  $\text{M}^+$  (568)].

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EXAMPLE 35

2-Ethoxycarbonyl-2-but-2-enyl (6R,7R)-7-[2-(2-Aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(S)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate

Sodium (6R,7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(S)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate, (0.35g) in 1-methyl-2-pyrrolidinone (4ml) was treated with a solution of ethyl (Z)-2-bromomethylbut-2-enoate, (0.16g) in 1-methyl-2-pyrrolidinone (1ml) and stirred at ambient temperature overnight. The solution was diluted with ethyl acetate and washed with water (3x), brine and then dried. After removal of solvent in vacuo, the residue was purified by flash chromatography on silica gel, eluting with 70, 90% ethyl acetate-hexane and then ethyl acetate. The title compound was obtained as a pale yellow foam (0.368g, 86%);  $\nu_{\text{max}}$  (CH<sub>2</sub>Cl<sub>2</sub>) 3480, 3389, 3320, 1781, 1726, 1682, 1606 and 1532cm<sup>-1</sup>;  $\delta_{\text{H}}$  (CDCl<sub>3</sub>) 1.30 (4H, t,  $\underline{J}$  7.1Hz, overlapping M), 1.66 (1H, m), 1.97 (3H, d,  $\underline{J}$  7.3Hz), 2.35 (1H, m), 3.33 and 3.64 (2H, ABq,  $\underline{J}$  18.7Hz), 3.88 (2H, m), 4.08 (3H, s), 4.22 (2H, q,  $\underline{J}$  7.1Hz), 4.93 (1H, m), 5.05 (3H, m), 5.80 (2H, br s, exch.), 5.99 (1H, dd,  $\underline{J}$  4.8, 9.0Hz, collapses to d,  $\underline{J}$  4.8Hz on exch.), 6.83 (1H, s), 7.21 (1H, q,  $\underline{J}$  7.3Hz) and 7.73 (1H, d,  $\underline{J}$  9.0Hz, exch.). [Mass spectrum: +ve ion (thioglycerol)  $\text{MH}^+$  (580)].

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In Vitro Biological DataMIC (µg/ml)

Example No.	Organism	
	E.coli (NCTC 1048)	S.aureus (Oxford)
1	0.50	1.00
3	2.00	1.00
5	0.50	0.25
7	0.50	1.00
9	1.00	0.50
13	1.00	4.00
17	1.00	2.00
18	16.00	1.00
19	4.00	2.00
21	0.25	8.00
22	8.00	0.25
24	0.12	1.00
27	4.00	1.00
28	>32	0.50

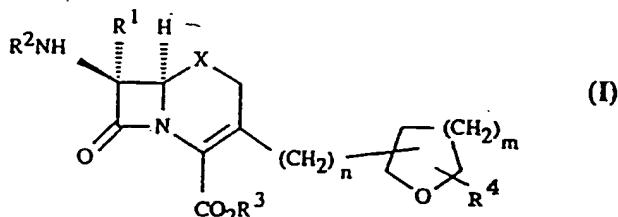
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Claims

1. A compound of formula (I) or a salt thereof:

5

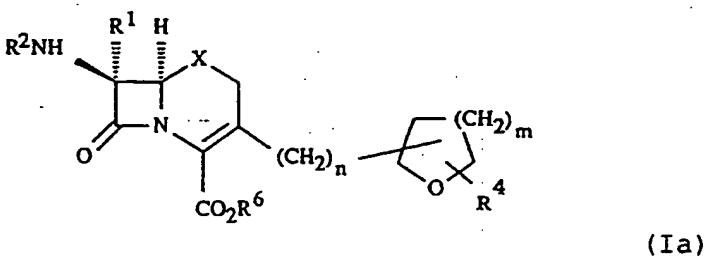


10

wherein

R<sup>1</sup> is hydrogen, methoxy or formamido;R<sup>2</sup> is an acyl group;CO<sub>2</sub>R<sup>3</sup> is a carboxy group or a carboxylate anion, or R<sup>3</sup> is a  
15 readily removable carboxy protecting group;R<sup>4</sup> represents up to four substituents selected from alkyl,  
alkenyl, alkynyl, alkoxy, hydroxy, halogen, amino,  
alkylamino, acylamino, dialkylamino, CO<sub>2</sub>R, CONR<sub>2</sub>, SO<sub>2</sub>NR<sub>2</sub>  
(where R is hydrogen or C<sub>1-6</sub> alkyl), aryl and heterocyclyl,  
20 which may be the same or different and wherein any R<sup>4</sup> alkyl  
substituent is optionally substituted by any other R<sup>4</sup>  
substituent; X is S, SO, SO<sub>2</sub>, O or CH<sub>2</sub>; m is 1 or 2; and n  
is 0.25 2. A compound as claimed in claim 1 having the formula  
(Ia):

30



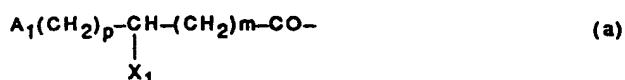
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wherein  $R^1$ ,  $R^2$ ,  $R^4$ ,  $m$ ,  $n$  and  $X$  are as defined with respect to formula (I) in claim 1 and the group  $CO_2R^6$  is  $CO_2R^3$  where  $CO_2R^3$  is a carboxy group or a carboxylate anion, or a pharmaceutically acceptable salt or in vivo hydrolysable ester thereof.

3. A compound as claimed in claim 1 or claim 2 wherein  $R^1$  is hydrogen.

10 4. A compound as claimed in claim 1,2 or 3 wherein  $R^2$  is an acyl group of formula (a) to (f):

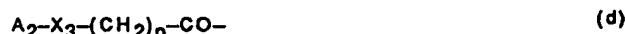
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wherein p is 0, 1 or 2; m is 0, 1 or 2; A<sub>1</sub> is C<sub>1-6</sub> alkyl, substituted C<sub>1-6</sub> alkyl, C<sub>3-6</sub> cycloalkyl, cyclohexenyl, cyclohexadienyl, an aromatic or heteroaromatic group; X<sub>1</sub> is a hydrogen or halogen atom, a carboxylic acid, carboxylic ester, sulphonic acid, azido, tetrazolyl, hydroxy, acyloxy, amino, ureido, acylamino, heterocyclamino, guanidino or acylureido group; A<sub>2</sub> is an aromatic or heteroaromatic group, a substituted alkyl group; or a substituted dithietane; X<sub>2</sub> is a -CH<sub>2</sub>OCH<sub>2</sub>-, -CH<sub>2</sub>SCH<sub>2</sub>- or alkylene group; X<sub>3</sub> is an oxygen 10 or sulphur atom; A<sub>3</sub> is an aryl or heteroaryl group; and A<sub>4</sub> is hydrogen, C<sub>1-6</sub>alkyl, C<sub>3-8</sub> cycloalkyl, C<sub>3-8</sub> cycloalkyl(C<sub>1-6</sub>)alkyl, C<sub>1-6</sub> alkoxy carbonyl(C<sub>1-6</sub>) alkyl, C<sub>2-6</sub> alkenyl, carboxy(C<sub>1-6</sub>)alkyl, C<sub>2-6</sub> alkynyl, aryl or C<sub>1-6</sub>alkyl substituted by up to three aryl groups.

15

5. A compound as claimed in claim 4 wherein A<sub>1</sub> is optionally substituted phenyl, X<sub>1</sub> is hydrogen or amino, A<sub>2</sub> is optionally substituted phenyl, X<sub>3</sub> is oxygen, A<sub>3</sub> is aminothiazolyl, aminothiadiazolyl or furyl, and R<sub>4</sub> is 20 hydrogen, C<sub>1-6</sub> alkyl, or carboxy C<sub>1-6</sub> alkyl.

6. A compound as claimed in any one of claims 1 to 5 wherein CO<sub>2</sub>R<sup>3</sup> is carboxy or a carboxylate anion or R<sup>3</sup> is t-butyl, 4-methoxybenzyl, diphenylmethyl, acetoxyethyl, 25 acetoxyethyl, pivaloyloxymethyl, propan-2-yloxycarbonyl-oxyethyl or 2-ethoxycarbonyl-but-2-enyl.

7. A compound as claimed in any one of claims 1 to 6 wherein the cyclic ether group bonded to the 3-position of 30 the cephalosporin nucleus is unsubstituted or unsubstituted by up to three substituents selected from C<sub>1-6</sub> alkyl, C<sub>1-6</sub> alkoxy, C<sub>1-6</sub> alkoxy carbonyl, C<sub>1-6</sub> alkanoyloxy C<sub>1-6</sub> alkyl or C<sub>1-6</sub> alkoxy C<sub>1-6</sub> alkyl.

35 8. A compound as claimed in any one of claims 1 to 7 wherein m is 1.

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9. A compound as claimed in any one of claims 1 to 8 wherein the cyclic ether group is a tetrahydrofuran-2-yl or a tetrahydropyran-2-yl group.

5 10. Sodium  $(6R, 7R)$ -7-[2-(2-Aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(RS)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate.

11. Pivaloyloxymethyl  $(6R, 7R)$ -7-[2-(2-Aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(RS)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate.

12. Sodium  $(6R, 7R)$ -7-[2-(2-Aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(RS)-tetrahydropyran-2-yl]-15 ceph-3-em-4-carboxylate.

13. Pivaloyloxymethyl  $(6R, 7R)$ -7-[2-(2-Aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(RS)-tetrahydropyran-2-yl]ceph-3-em-4-carboxylate.

20 14.  $(6R, 7R)$ -7-[2-(2-Aminothiazol-4-yl)-2-(Z)-hydroxyiminoacetamido]-3-[(RS)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylic acid.

25 15. Sodium  $(6R, 7R)$ -7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(S)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate.

16. Pivaloyloxymethyl  $(6R, 7R)$ -7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(S)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate.

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17. Sodium (6R, 7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(R)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate.

5 18. Pivaloyloxymethyl (6R, 7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(R)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate.

10 19. Diphenylmethyl (6R, 7R)-7-phenylacetamido-3-[(RS)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate.

20. Sodium (6R, 7R)-7-[2-(2-Aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(RS)-tetrahydrofuran-3-yl]ceph-3-em-4-carboxylate.

15

21. Acetoxyethyl (6R, 7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(S)-tetrahydrofuran-2-yl]-ceph-3-em-4-carboxylate.

20 22. Sodium (6R, 7R)-7-[2-(2-Aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-(5-methoxymethyltetrahydrofuran-2-yl)ceph-3-em-4-carboxylate

23. Sodium (6R, 7R)-7-[2-(2-Aminothiazol-4-yl)-2-(Z)-pent-2-enamido]-3-[(S)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate.

24. Sodium (6R, 7R)-7-[2-(2-Aminothiadiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(S)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate.

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25. (RS)-1-Acetoxyethyl (6R,7R)-7-[2-(2-Aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(S)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate.

5 26. (6R,7R)-7-[2-(2-Aminothiazol-4-yl)-2-(Z)-carboxy-methoxyiminoacetamido]-3-[(RS)-tetrahydrofuran-2-yl]-ceph-3-em-4-carboxylic acid disodium salt.

27. Sodium (6R,7R)-7-[(R)-2-Amino-2-(4-hydroxyphenyl)-acetamido]-3-[(S)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate.

28. Sodium (1S,6R,7R)-7-[2-(2-Aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(S)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate-1-oxide.

29. Sodium 7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxy-iminoacetamido]-3-(tetrahydrofuran-2-yl)-1-carba-1-dethia-ceph-3-em-4-carboxylate.

20

30. Sodium (6R,7R)-7-[2-(2-Aminothiazol-4-yl)-2-(Z)-methoxy-iminoacetamido]-3-[(S)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate-1,1-dioxide.

25 31. (RS)-1-(Propan-2-yl)oxycarbonyloxyethyl (6R,7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(S)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate.

32. Sodium (6R,7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(5R,2SR)-5-methyltetrahydrofuran-2-yl]ceph-3-em-4-carboxylate.

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33. Sodium (6R, 7R)-7-[2-(furan-2-yl)-2-(Z)-methoxyiminoacetamido]-3-[(S)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate.

5 34. Sodium (6R, 7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(S)-5,5-dimethyltetrahydrofuran-2-yl]ceph-3-em-4-carboxylate.

10 35. Sodium (6R, 7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-(5-methoxycarbonyltetrahydrofuran-2-yl)ceph-3-em-4-carboxylate.

15 36. Sodium (6R, 7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[3-methyltetrahydrofuran-2-yl]ceph-3-em-4-carboxylate.

37. 2-Ethoxycarbonyl-(Z)-but-2-enyl (6R, 7R)-7-[2-(2-aminothiazol-4-yl)-2-(Z)-methoxyiminoacetamido]-3-[(S)-tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate.

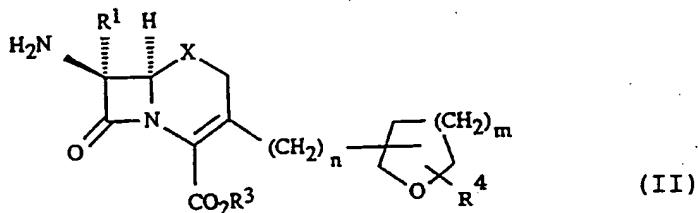
20 38. A compound of formula (I) as defined in claim 1 substantially as hereinbefore described with reference to the preparative examples.

25 39. A process for the preparation of a compound of formula (I) as defined in any one of claims 1 to 28 which process comprises:

(a) treating a compound of formula (II) or a salt thereof:

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wherein  $R^1$ ,  $CO_2R^3$ ,  $R^4$ ,  $m$ ,  $n$ , and  $X$  are as hereinbefore defined with respect to formula (I) in claim 1, wherein any reactive group may be protected, and wherein the amino group is optionally substituted with a group which permits 5 acylation to take place, with an N-acylating derivative of an acid of formula (III):

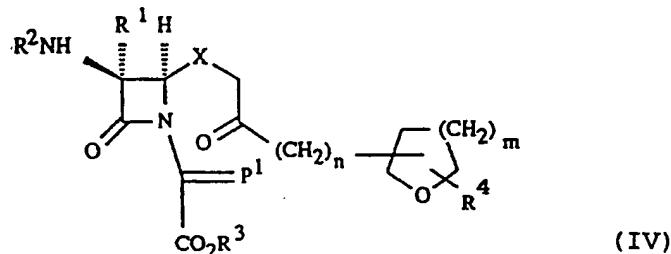


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wherein  $R^2$  is as hereinbefore defined with respect to formula (I) in claim 1 and wherein any reactive group may be protected; or

15 (b) cyclising a compound of formula (IV):

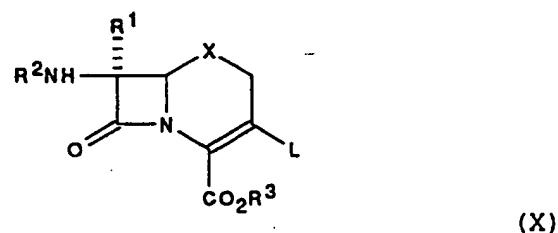
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wherein  $X$ ,  $R^1$ ,  $R^2$ ,  $R^4$ ,  $m$ ,  $n$  and  $CO_2R^3$  are as hereinbefore defined with respect to formula (I) in claim 1 and  $P'$  is a 25 phosphorus residue; or

(c) treating a compound of formula (X):

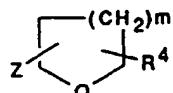
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35 wherein  $R^1$ ,  $R^2$ ,  $CO_2R^3$  and  $X$  are as hereinbefore defined with respect to formula (I) in claim 1, and  $L$  is a leaving group, with a compound of formula (XI):

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(XI)

wherein Z is an organo-cuprate group and R<sup>4</sup> and m are as hereinbefore defined with respect to formula (I) in claim 1;

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and thereafter, if necessary or desired, carrying out one of the following steps:

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- i) removing any protecting groups;
- ii) converting the group CO<sub>2</sub>R<sup>3</sup> to a different group CO<sub>2</sub>R<sup>3</sup>;
- iii) converting the group R<sup>2</sup> to a different group R<sup>2</sup>;
- iv) converting the group X to a different group X;
- v) converting the product into a salt.

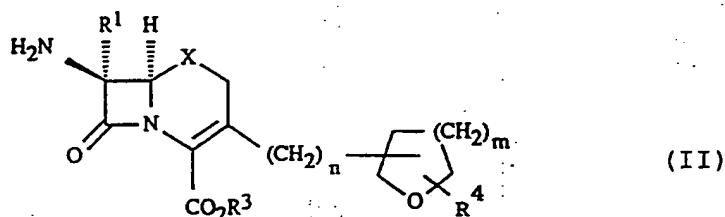
40. A process for the preparation of a compound of formula (I) substantially as hereinbefore described in the preparative Examples.

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41. A compound of formula (II) or a salt thereof:

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(II)

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wherein  $R^1$   $CO_2R^3$ ,  $R^4$ ,  $x$ ,  $m$  and  $n$  are as hereinbefore defined with respect to formula (I) in claim 1.

42.  $t$ -Butyl  $6R, 7R$ -7-Amino-3-(tetrahydrofuran-2-yl)-5-ceph-3-em-4-carboxylate.

43.  $t$ -Butyl  $(6R, 7R)$ -7-Amino-3-[ $(RS)$ -tetrahydropyran-2-yl]ceph-3-em-4-carboxylate.

10 44. 4-Methoxybenzyl  $(6R, 7R)$ -7-amino-3-(tetrahydrofuran-2-yl)ceph-3-em-4-carboxylate.

45. Pivaloyloxymethyl  $(6R, 7R)$ -7-amino-3-(tetrahydrofuran-2-yl)ceph-3-em-4-carboxylate.

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46.  $t$ -Butyl  $(6R, 7R)$ -7-Amino-3-[ $(RS)$ -tetrahydrofuran-3-yl]ceph-3-em-4-carboxylate.

47. Acetoxyethyl  $(6R, 7R)$ -7-amino-3-[ $(S)$ -tetrahydrofuran-2-yl]ceph-3-em-4-carboxylate.

48. 4-Methoxybenzyl  $(6R, 7R)$ -7-Amino-3-(5-methoxymethyl-tetrahydrofuran-2-yl)ceph-3-em-4-carboxylate.

25 49. 4-Methoxybenzyl  $(6RS, 7SR)$ -7-amino-3-(tetrahydrofuran-2-yl)-1-carba-1-dethiaceph-3-em-4-carboxylate.

50. 4-Methoxybenzyl  $(6R, 7R)$ -7-amino-3-(5-methyl-tetrahydrofuran-2-yl)ceph-3-em-4-carboxylate.

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51. A compound of formula (II) as defined in claim 41 substantially as hereinbefore described with reference to the preparative Examples.

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52. A pharmaceutical composition comprising a compound of formula (Ia) as defined in claim 2 or a pharmaceutically acceptable salt or in vivo hydrolysable ester thereof, and a pharmaceutically acceptable carrier.

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53. A pharmaceutical composition as claimed in claim 52 further comprising a  $\beta$ -lactamase inhibitor.

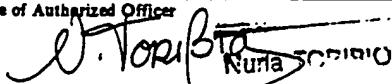
54. A compound of formula (Ia) or a pharmaceutically acceptable salt or in vivo hydrolysable ester thereof as defined in claim 2, for use as a therapeutic agent.

55. A method of treating bacterial infections in humans and animals which comprises administering a therapeutically effective amount of a compound of formula (Ia) or a pharmaceutically acceptable salt or in vivo hydrolysable ester thereof, as defined in claim 2, to a human or animal.

56. The use of a compound of formula (Ia) or a pharmaceutically acceptable salt or in vivo hydrolysable ester thereof, as defined in claim 2, for the manufacture of a medicament for the treatment of bacterial infections.

# INTERNATIONAL SEARCH REPORT

International Appl. No. PCT/GB 91/01228

<b>I. CLASSIFICATION OF SUBJECT MATTER</b> (if several classification symbols apply, indicate all) <sup>6</sup>			
According to International Patent Classification (IPC) or to both National Classification and IPC			
Int.C1.5	C 07 D 501/20	C 07 D 501/18	C 07 D 463/00
A 61 K 31/545	A 61 K 31/435		
<b>II. FIELDS SEARCHED</b>			
Minimum Documentation Searched <sup>7</sup>			
Classification System	Classification Symbols		
Int.C1.5	C 07 D 501/00 C 07 D 463/00		
Documentation Searched other than Minimum Documentation to the Extent that such Documents are Included in the Fields Searched <sup>8</sup>			
<b>III. DOCUMENTS CONSIDERED TO BE RELEVANT<sup>9</sup></b>			
Category <sup>10</sup>	Citation of Document, <sup>11</sup> with indication, where appropriate, of the relevant passages <sup>12</sup>	Relevant to Claim No. <sup>13</sup>	
A	US,A,3962223 (JACQUES MARTEL) 8 June 1976, see abstract ---	1-56	
A	FR,A,2197573 (ROUSSEL UCLAF) 29 March 1974, see pages 22-29; claims; formula page ---	1-56	
A	FR,A,2183892 (HOECHST) 21 December 1973, see pages 1-19; claims (cited in the application) ---	1-56	
P,A	EP,A,0395219 (BEECHAM) 31 October 1990, see the whole specification -----	1-56	
<b>IV. CERTIFICATION</b>			
Date of the Actual Completion of the International Search	Date of Mailing of this International Search Report		
08-10-1991	09.12.91		
International Searching Authority EUROPEAN PATENT OFFICE	Signature of Authorized Officer  Nuria Toribio		

## FURTHER INFORMATION CONTINUED FROM THE SECOND SHEET

V.  OBSERVATION WHERE CERTAIN CLAIMS WERE FOUND UNSEARCHABLE <sup>1</sup>

This International search report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1.  Claim numbers 55 because they relate to subject matter not required to be searched by this Authority, namely:  
see rule 39.1(iv)

2.  Claim numbers because they relate to parts of the International application that do not comply with the prescribed requirements to such an extent that no meaningful International search can be carried out, specifically:

The search has been systematically performed for the compounds of formula I and II as far as X represents -CH - or S and its oxides (the only examples described)

3.  Claim numbers because they are dependent claims and are not drafted in accordance with the second and third sentences of PCT Rule 6.4(a).

VI.  OBSERVATIONS WHERE UNITY OF INVENTION IS LACKING <sup>2</sup>

This International Searching Authority found multiple inventions in this International application as follows:

- As all required additional search fees were timely paid by the applicant, this International search report covers all searchable claims of the International application
- As only some of the required additional search fees were timely paid by the applicant, this International search report covers only those claims of the International application for which fees were paid, specifically claims:
- No required additional search fees were timely paid by the applicant. Consequently, this International search report is restricted to the invention first mentioned in the claims; it is covered by claim numbers:
- As all searchable claims could be searched without effort justifying an additional fee, the International Searching Authority did not invite payment of any additional fee.

## Remark on Protest

The additional search fees were accompanied by applicant's protest.  
 No protest accompanied the payment of additional search fees.

**ANNEX TO THE INTERNATIONAL SEARCH REPORT  
ON INTERNATIONAL PATENT APPLICATION NO.**

GB 9101228  
SA 49650

This annex lists the patent family members relating to the patent documents cited in the above-mentioned international search report. The members are as contained in the European Patent Office EDP file on 05/11/91. The European Patent Office is in no way liable for these particulars which are merely given for the purpose of information.

Patent document cited in search report	Publication date	Patent family member(s)		Publication date
US-A- 3962223	08-06-76	FR-A-	2166620	17-08-73
		FR-A-	2166621	17-08-73
		FR-A-	2166622	17-08-73
		FR-A-	2166623	17-08-73
		FR-A-	2165790	10-08-73
		FR-A-	2197573	29-03-74
		AT-B-	324554	10-09-75
		AU-B-	473686	01-07-76
		AU-A-	5067973	04-07-74
		BE-A-	793448	28-06-73
		CA-A-	999850	16-11-76
		CA-A-	994799	10-08-76
		CA-A-	1019321	18-10-77
		CH-A-	577519	15-07-76
		CH-A-	577516	15-07-76
		CH-A-	568328	31-10-75
		CH-A-	563398	30-06-75
		DE-A,C	2300009	12-07-73
		GB-A-	1423741	04-02-76
		GB-A-	1423742	04-02-76
		JP-C-	1206682	11-05-84
		JP-A-	55149228	20-11-80
		JP-B-	58037297	15-08-83
		NL-A-	7300031	03-07-73
		SE-B-	415564	13-10-80
		SE-B-	426816	14-02-83
		SE-A-	7606177	01-06-76
		SE-B-	417824	13-04-81
		SE-A-	7606178	02-06-76
		US-A-	4016158	05-04-77
		GB-A-	1423743	04-02-76
FR-A- 2197573	29-03-74	AT-B-	324554	10-09-75
		BE-A-	793448	28-06-73
		CH-A-	577519	15-07-76
		CH-A-	577516	15-07-76
		CH-A-	568328	31-10-75
		CH-A-	563398	30-06-75
		DE-A,C	2300009	12-07-73

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ON INTERNATIONAL PATENT APPLICATION NO.**

GB 9101228  
SA 49650

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Patent document cited in search report	Publication date	Patent family member(s)	Publication date
FR-A- 2197573		GB-A- 1423741 GB-A- 1423743 GB-A- 1423742 NL-A- 7300031 SE-B- 415564 SE-B- 426816 SE-A- 7606177 SE-B- 417824 SE-A- 7606178 US-A- 3962223 US-A- 4016158	04-02-76 04-02-76 04-02-76 03-07-73 13-10-80 14-02-83 01-06-76 13-04-81 02-06-76 08-06-76 05-04-77
FR-A- 2183892	21-12-73	DE-A- 2222140 DE-A- 2316541 AT-B- 324559 AU-B- 469255 AU-A- 5516273 BE-A- 799200 CH-A- 579574 GB-A- 1385831 JP-A- 49061189 NL-A- 7305936 US-A- 3920640	22-11-73 24-10-74 10-09-75 05-02-76 14-11-74 07-11-73 15-09-76 05-03-75 13-06-74 07-11-73 18-11-75
EP-A- 0395219	31-10-90	AU-A- 5232190 JP-A- 2286680	04-10-90 26-11-90